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ELECTRONYSTAGMOGRAPHIC FINDINGS IN MULTIPLE SCLEROSIS

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(Received April 7 1972)

Abstract. Fifty MS cases were studied both clinically and by neuro-otological methods. MS diagnosis was verified using both Schumacher's criteria and cerebrospinal fluid (CSF) findings. Seventy-two per cent of the subjects were studied within the first 3 years after the appearance of the first possible anamnestic symptoms of the disease. Only 3 of the patients had suffered from MS for more than 13 years. Special emphasis was paid to those cases which came in for study at a relatively early stage of the disease. Either spontaneous or positional nystagmus was found in 60% of all cases. No significant hearing disturbances were found in our material. The present results suggest that electronystagmography and other neuro-otological techniques provide a valuable aid in the early differential diagnosis of MS because of their sensitivity and the close relationship between predilection sites of MS and those structures which contribute to the maintenance of balance.

Multiple sclerosis (MS) is the most common of the demyelinating diseases of the central nervous system. According to Panelius (1969) its prevalence in the southwestern part of Finland is 35.4 for 100 000 inhabitants. It is a well established fact that the disease lesions in multiple sclerosis (MS) are diffusely scattered throughout the white matter but at the same time there are also some predilection areas in the brain which often show clinical signs earlier than others. Such regions are, for example, the brain stem and optic nerves where slight morphological changes can actually lead to profound clinical manifestations (Lumsden, 1970). Although MS may begin with almost any neurological symptom vertigo and nystagmus are often mentioned as preliminary manifestations before verification of diagnosis in clinical studies (Müller 1949).

McAlpine et al. (1965) concludes that ves-

tibular and auditory findings in MS are variable. The contradictory findings of different authors may be due to the fact that some investigators include among brain stem manifestations only those signs which are found at routine neurological examination. Differing results may therefore be expected in studies where more advanced methods, aimed at testing vestibular responses and nystagmus, are used. It must also be borne in mind that these findings are very difficult to interpret and it is therefore essential for clinical purposes to follow the changes in electronystagmographic findings during the various stages of the disease so that their significance as a diagnostic aid in demonstrating brain stem lesions may be evaluated, especially during the early stages of MS.

This study contains a report on the type and frequency of various neuro-otological findings in diagnosed MS cases. Special emphasis is given in this material to those MS cases who came under our care at a relatively early stage of the disease because it is often difficult to demonstrate changes at routine neurological examination.

MATERIAL AND METHODS

Patient material

The material consists of 50 patients who underwent neuro-otological examination for various reasons. MS diagnosis was made on the basis of Schumacher's criteria (Schumacher et al., 1965) for all patients. The major criteria in

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tion-determined in 10 cases (Nylén's type II) and irregular direction-changing in 2 cases (Nylén's type III). Nystagmus was spontaneous in 17 cases. It is important to stress that hearing tests were normal. This shows that the peripheral part of the labyrinth system would give normal reactions by caloric irrigations. Despite this assumption it was found that caloric irritability was decreased in 18 cases, showing that MS affects the first neuron in the vestibular pathways more than neurons mediating purely auditive information. Directional preponderance was found in 14 cases.

The ocular fixation index was higher than 50% in 22 patients. (In other words the fixation did not suppress the caloric nystagmus as it would in normal subjects and those patients probably have a central lesion in the vestibular pathway.) Although all patients were asked to perform mental arithmetic in order to keep them awake dysrhythmic caloric reaction patterns were observed in 16 cases. This also suggests the presence of a central lesion in vestibular pathways either in the brain stem or at a higher central level.

DISCUSSION

It is well established that clinical history relapses, fluctuating course, disseminated neuro-

Table I. Age distribution, age at onset and time between onset of first symptoms and neuro-otological examination

Total number of multiple sclerosis cases is 50 (23 males, average age of onset 34.3 years, 27 females average age of onset 32.5 years)

Age groups	Age distribution (no. of cases)	Age at the age of onset examination (no. of cases)	Duration of illness before neuro-otological examination (years)	
15-25	4	7	0-2	14
26-35	17	19	2-3	22
36-45	23	22	3-4	5
46-55	4	2	4-5	2
56-65	2	0	5-6	1
			7-9	1
			over 13	5

Table II. Neurological deficiency symptoms (cerebellar and brain stem symptoms pyramidal signs ocular muscle paresis and intranuclear ophthalmoplegia facial abnormalities) and positive neuro-otological symptoms (positional nystagmus according to Nylén and spontaneous nystagmus)

Total number of patients was 50

	No. of positive findings	Unilateral	Bi-lateral
Dysmetria	32	5	27
Dysidiadochokinesis	44	3	17
Ataxia	20		
Extensor plantar response	38	8	30
Ocular muscle paresis of intranuclear ophthalmoplegia	27		
Facial weakness or facial paresis	10	9	1
Positional nystagmus			
Nylén type I	4		
Nylén type II	10		
Nylén type III	2		
Spontaneous nystagmus	17		
Directional preponderance	14		
Hypocapnibility in caloric test	18		
OPI higher than 50 percent	22		

logical signs and positive CSF findings are all helpful in making MS diagnoses. There are, however cases where differential diagnostic problems are extremely difficult. The present results show that involvement of the vestibular system and its central connections already exists at an early stage of the disease. We do not of course assume that positive findings in neuro-otological examination are alone sufficient to conclude that a patient is suffering from MS. However when there are certain other scattered signs in the neurological examination, such as transient spinal level symptoms, optic neuritis, slight monoparesis, paraparesis and possibly also other findings suggesting that neurological lesions are outside the brain stem we may say that electronystagmographic examination is of real value in revealing a dysfunction in the vestibular system and its central connections. Detailed neuro-otological ex-

amination seems to reveal disease affections in the brain stem earlier and more selectively than routine neurological examination. The significance of such things as facial asymmetry and nystagmus in different patient series are very difficult to evaluate and much will depend on the experience of the clinician in question. Facial weakness was found in 5% of the cases in our material. The corresponding figures in Kurtzke's material (1970 a) are 2.6% in Müller's material (1949) 11.0% and in Abb's material (Abb & Schaltenbrand, 1956) 14.4%. Nystagmus is one of the most common findings in MS. According to Abb & Schaltenbrand (1956) it is present in 40% of all cases and in Müller's material (1949) it was found in 17%. In our material the corresponding percentage was 60. The differences in these findings can easily be explained on the basis of the differences in the age distribution of the material and differences in the duration of the disease.

Vertigo is caused by a very complicated pattern of functional disturbances which include both complex peripheral vestibular and central functional lesions due to various disease processes. Vertigo is found frequently in the clinical history of MS patients and it seems to be one of the most common early manifestations (Müller 1949 Hahn et al. 1959). Seen against this background the results in the present study are very interesting for differential diagnostic purposes. They show that abnormalities in hearing are found only occasionally. If this can be confirmed for wider MS material, it will also help in the differential diagnosis of MS especially if at the same time the common brain stem symptoms caused by cerebrovascular lesions in the vertebro-basillary region are taken into account. As is well known, vascular lesions often cause disturbances in hearing which can be demonstrated by audiometry. The same is true of such tumours as acoustic neurinoma where the clinical picture may resemble brain stem multiple sclerosis. The differential diagnosis of cerebellar lesions and brain stem affections which are caused by various tumours are a difficult

clinical problem and one must therefore always be extremely cautious when drawing conclusions. Exclusive neuroradiological examinations are often necessary in cases where there is a suspected clinical case history with increase of CSF proteins and positive neuro-otological findings, although no signs of increased intracranial pressure are demonstrable. From a clinical point of view vestibular neuronitis with acute onset is extremely difficult to distinguish from early MS because if in the former disease the question is one of brain stem encephalitis as is sometimes thought CSF findings may be sparse and very similar to those found in MS. In such cases, however, electronystagmography will provide valuable additional information. In vestibular neuronitis it can be demonstrated by ENG that the disturbance is due to peripheral lesion and such lesions are not often seen in MS. Some authors, especially in early literature, have described hearing abnormalities in MS. The results derived from the present material, where MS diagnoses were established on the basis of generally accepted international criteria and where audiometry was also used as a differential diagnostic aid show that before CSF findings are included in MS work there may have been a number of cases giving an "atypical" picture of MS in which the diagnoses were wrong and loss of hearing due to some other reason. It is of course possible that in some MS cases central hearing loss may occur. The present material included five cases in which the duration of disease was more than 13 years in none of them, however, was it possible to show any significant hearing abnormality. Present results therefore suggest that if central hearing abnormalities are found in detailed neuro-otological examination in an early stage of the disease process some other diagnosis than MS should be considered. Such neurological symptoms as dysarthria, intention tremor, dysidiadocho-kinesia and ataxia are usually mentioned as symptoms of MS (Kurtzke 1970 b). These were also found in our material but it seems that positive electronystagmographic findings are a better indicator

of early stages of MS. In other words vestibular dysfunction in one form or another without defect in hearing together with slight or evident pathology in CSF immunoglobulins should lead the clinician to consider MS as an alternative diagnostic possibility even when there are no other clearly identifiable neurological signs. In such cases a continuous follow up of the patient, with the help of CSF tests and electronystagmographic techniques may provide the clinician with a valuable aid in what is often and exceedingly difficult diagnostic problem to demonstrate brain stem lesions.

ZUSAMMENFASSUNG

60 Fälle von MS wurden sowohl mit klinischen als neuro-otologischen Methoden untersucht. MS-Diagnose wurde durch Anwendung von sowohl Schumachers Kriterium als cerebrospinale Flüssigkeitsuntersuchung verifiziert. 72% von den Subjekten wurden innerhalb der ersten 3 Jahre nach Erscheinung der ersten wahrscheinlichen anatomischen Krankheitssymptome studiert. Nur fünf Patienten hatten an MS mehr als 13 Jahre gelitten. Besonders große Bedeutung wurde denjenigen Fällen gegeben, die in einem relativ frühen Stadium der Krankheit zur Untersuchung einkamen. Entweder spontanes oder posturales Nystagmus war in 60% aller Fälle zu finden. Keine signifikative Hornorumpen wurden in unserem Material gefunden.

Die vorliegenden Resultate geben an, daß Elektornystagmus und übrige neuro-otologische technische Methoden wertvolle Hilfe in der frühen differentiellen Diagnose von MS infolge ihrer Empfindlichkeit und nahe Verwandtschaft zwischen dem Prüfobjekts Gebiet von MS und denjenigen Strukturen, die an der Aufrechterhaltung der Balance teilnehmen, geben.

REFERENCES

- Abb, L. & Schaltenbrand, G. 1956. Statistische Untersuchungen zum Problem der multiplen Sklerosen. II. Mitteilung. Das Krankheitsbild der multiple Sklerose. *Deutscher Z. Nervenheilk.* 174 199.
- Demarez, J. P. & Ledoux, A. 1970. Automatic fixation mechanisms and vestibular stimulation. *Advances in Oto-Rhino-Laryngology* (ed. L. Rödel). Karger, Basel.
- Hahn, R., Scarzella, R. & Zarakda, A. 1959. Lesame vestibulaire nella sclerosi multipla. *Atterva Otorhinolaryng* 9 235.
- Kurtzke, J. F. 1970 a. Neurologic impairment in multiple sclerosis and disability status scale. *Acta Neurol Scand* 46 493.
- 1970 b. Diagnosis and differential diagnosis of multiple sclerosis. *Acta Neurol Scand* 46 484.
- Lumden, C. E. 1970. Neuropathology of multiple sclerosis. In *Handbook of clinical neurology* (ed. P. Vinken & G. W. Bruyn), p. 17. North-Holland, Amsterdam.
- McAlpine, D., Lumden, C. E. & Acheson, E. D. 1965. *Multiple sclerosis* p. 94. Livingstone, Edinburgh and London.
- Müller, R. 1949. Studies on disseminated sclerosis with special reference to symptomatology course and prognosis. *Acta Med Scand Suppl.* 22.
- Panelius, M. 1969. Studies on epidemiological, clinical and etiological aspects of multiple sclerosis. *Acta Neurol Scand Suppl.* 39.
- Schumacher, G. A., Berre, G., Kibler, R. F., Harland, L. T., Kurtzke, J. F., McDowell, F., Nagler, B., Sibley, W. A., Tourtellotte, W. W. & Willmon, T. L. 1965. Problems of experimental trials of therapy in multiple sclerosis. Export by the panel on the evaluation of experimental trials of therapy in multiple sclerosis. *Ann NY Acad Sci* 122 552.

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SEPARATE RECORDING OF THE MOVEMENTS OF THE HUMAN EYES DURING PARALLEL SWING TESTS

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(Received February 28, 1972)

Abstract In parallel swing tests different eye tracks were recorded. The eye movements of both eyes do not always seem to be related to one another. In some positions only one eye was moving. Sometimes the eye movements showed double the swing frequency. For this phenomenon a possible explanation is given.

In the literature concerning parallel swing it is generally accepted that both eyes react completely identically upon linear acceleration stimuli. In reality however the eye movements have practically never been recorded separately. Only in rabbits have separate eye movements been recorded, and under certain conditions different eye tracks have been obtained.

The present tests were made in an effort to determine whether or not the two human eyes follow an exactly identical pattern of movements when being stimulated on the parallel swing.

METHOD

The parallel swing used in these investigations had a period of 3.3 sec, and the amplitude used was about 30 cm. In order to avoid minor shiftings of the skin of the head with respect to the skull during the swinging process, on the parallel swing a second small parallel swing was mounted on which the test subjects rested their heads. The length of the cords of this second swing was 50 cm (Fig. 1).

Because the mass of the head was much greater than that of the swing on which it

rested, every possible movement was definitely prevented. This is very important, since minor shiftings of the electrodes may result in rather large signals which may easily lead to false interpretations. For the recording of the eye movements seven electrodes were used which were fixed in position as sketched (Fig. 2).

An eighth electrode was fixed on the wrist. Recordings were made as follows. Right eye: D-E, called "vertical" in text, $(A-C) + \beta(D+E)$, called "horizontal" in text. Left eye: F-G called "vertical" $(C-B) + \gamma(F-G)$ called "horizontal".

In the test procedure the gain factors β and γ were adjusted in such a way that vertical eye movements were no longer perceptible on the horizontal recordings. This effect was nearly always achieved.

In total 22 subjects have been tested on the swing. They were moved lengthwise in the following positions: 1) lying on the back, 2) lying on the right side, and 3) lying on the left side. Each subject was instructed to keep his eyes closed and to assume a reverie state as far as possible.

RESULTS

Lying on the back Sinusoidal horizontal eye movements were observed in 6 subjects. Out of these 6, 2 showed a sinusoid with double the swing frequency (Fig. 3). Attention is

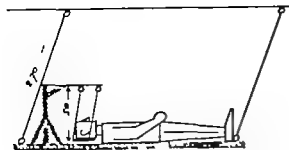


Fig 1



Fig 2

drawn to the opposite direction of the movements. In two other cases vertical identical eye movements were observed.

Lying on the right side: Only in 1 subject were vertical eye movements observed, whereas 15 subjects showed horizontal sinusoids. In this group 6 subjects were found in which only the right eye produced a sinusoidal pattern (Fig. 4).

Lying on the left side: In 14 subjects, horizontal sinusoids were observed. Six subjects

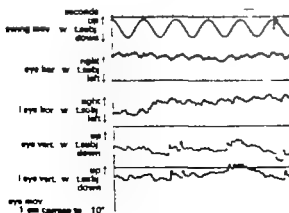


Fig 3

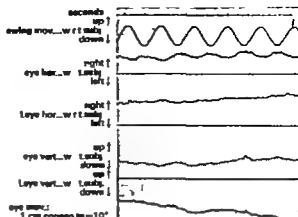


Fig 4

moved only the left eye. Moreover 11 subjects produced also vertical movements with the left eye (Fig. 5).

In all cases the phase differences between the eye movements and the swing movements varied among the subjects from -180 to $+180$.

A remarkable case is that of a man with right-sided labyrinthine dysfunction. After swinging on the left side it was observed that, while lying on the right side before swinging, the right eye produced sinusoids with about the same frequency as the swing frequency (Fig. 6).

DISCUSSION

In some cases the sinusoidal eye movements began only after 4 minutes of constant ampli-

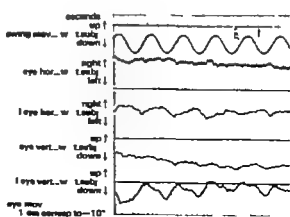


Fig 5

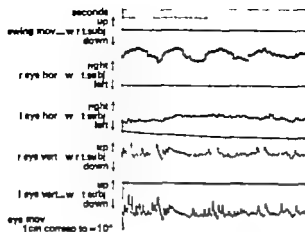


Fig 6

tude swinging. It is concluded, therefore that faulty measurement techniques were definitely not the source of the signals measured.

There are some indications that a vestibular system under pressure reacts more intensively. The pressure on the underlying labyrinth is approximately 13 cm H₂O higher than on the upper labyrinth. The eye movements are known to be more closely linked to the homolateral labyrinth. This is a possible explanation for the larger excursions of the underlying eye.

The occurrence of double the swing frequency may be the result of the vertical oscillations of the swing. An analysis shows that if we portray the otoliths schematically as in Fig. 7 they are subjected to the following accelerations as a result of the swing movements.

If $\alpha(t) = \alpha_m \cos \omega t$ we use the following approximations because α_m is very small

$$\sin(\alpha_m \cos \omega t) \approx \alpha_m \cos \omega t$$

$$\cos(\alpha_m \cos \omega t) \approx 1 - \frac{1}{2} \alpha_m^2 \cos^2 \omega t$$

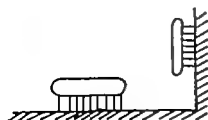


Fig 7

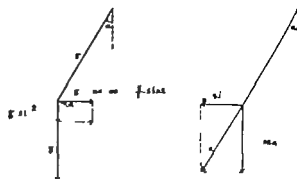


Fig 8

$$\begin{aligned} \text{If } \omega_h = \frac{dx}{dt} = \omega_m \sin \omega t \rightarrow a_x = \omega_h^2 r = \left(\frac{dx}{dt}\right)^2 r^2 \\ = \omega^2 \alpha_m^2 r^2 \sin^2 \omega t \end{aligned}$$

$$\text{Hor acc: } -g/2 \sin 2\alpha - a_x \sin \alpha = -g \alpha_m \cos \omega t - r \omega^2 \alpha_m^2 \sin^2 \omega t \cos \omega t$$

Because $\alpha = 0.12 \text{ rad}$, $\omega = 2 \text{ Hz}$, $r = 2.5 \text{ m}$, the acceleration is $-1.2 \cos \omega t - 0.02 \sin^2 \omega t \cos \omega t$

$$\begin{aligned} \text{Vert acc: } -g \sin^2 \alpha + a_x \cos \alpha = -g \alpha_m^2 \\ \cos^2 \omega t + r \omega^2 \alpha_m^2 \sin^2 \omega t (1 - \alpha_m^2 \cos^2 \omega t) \\ \rightarrow -0.14 \cos^2 \omega t + 0.14 \sin^2 \omega t = -0.14 \cos 2\omega t \end{aligned}$$

This maximal vertical acceleration of 14 cm sec^{-2} exceeds the generally agreed minimum perceptible acceleration.

CONCLUSIONS

1 For the swing acceleration of 120 cm/sec^2 not all the subjects showed a response.

2 In the side positions the response was not the same for both eyes. In cases where only one eye responded it was always the lower eye.

3 Several instances occurred in which the response was of twice the frequency of the swing. Dynamic mechanical analysis indicates that this response may be the result of the vertical accelerations of the parallel swing.

ACKNOWLEDGMENT

We thank Miss Anneke Matthijs for her administrative assistance and Mr S G Mey and Mr A. M. Sad for their valuable technical help.

ZUSAMMENFASSUNG

In Parallelschaukel Tests sind ungleiche Augenbewegungen registriert worden. Die Bewegungen von beiden Augen scheinen nicht immer miteinander übereinzustimmen. Es gab Positionen, in denen nur ein Auge sich bewegte. Manchmal war die Frequenz der Augen-

bewegung zweimal so gross wie die der Schaukel. Für dieses Phänomen ist eine mögliche Erklärung gegeben worden.

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REVISED NORMATIVE STANDARDS OF PERFORMANCE OF MEN ON A QUANTITATIVE ATAXIA TEST BATTERY

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(Received April 6, 1972)

Abstract Revised normative standards of performance are presented, of men, ages 16-60 years, on a quantitative ataxia test battery in terms of five new chronological age groupings based on a sample of 1 055 normal men. Very nearly similar sensitivity of all battery subtests to aging influences was observed, which implies a unitary sensitivity of the battery to such influences on the complex postural equilibrium functions sampled by it. Results tentatively suggest that the progressive nature of negative aging influences along the life span sampled become apparent several years earlier than reported previously. The new normative standards should allow more precise laboratory and clinical applications of the test battery which has proved to be useful particularly in the vestibular physiological and neuro-otological assessment of postural equilibrium-disequilibrium, including spontaneous and induced vestibular ataxia. The test battery has other clinical and research applications.

Several years ago age-influenced normative standards of performance on a new quantitative ataxia test battery designed initially for assessment of vestibular ataxia (Fregly in press; Graybiel & Fregly 1966)¹ were published (Fregly & Graybiel, 1970; Graybiel &

Fregly 1966). To control for the observed negative influence of chronological aging on scores, the normative standards were presented in relation to several chronological age groupings. Meanwhile, many additional individuals—almost entirely men—were tested, and new indications of aging influences on test scores came to light. Accordingly normative standards were revised in terms of new chronological age groupings, incorporating the additional samples of normal men tested and new 5th percentile and below cut-off scores relative to an arbitrary definition of frank ataxia (Fregly & Graybiel, 1970; Fregly et al. 1971). This up-dated information is presented and discussed to allow more precise laboratory and clinical applications of the test battery.

PROCEDURE

Subjects

The sample of 1 055 normal men, 16 to 60 years of age included all those physically fit who had been tested since standardization of the short version of the "rail battery" and the "floor battery" of ataxia tests (Fregly in press; Fregly et al. 1971; Graybiel & Fregly 1966). Occupational groups included experienced and student military and civilian aviators; Army and Navy enlisted and civilian college student volunteer research subjects; Navy deep-sea divers and miscellaneous active and retired

This research was conducted under the sponsorship of the Bureau of Medicine and Surgery work unit NRO-41 01 01-0120BSFG.

Opinions or conclusions contained in this report are those of the authors and do not necessarily reflect the views or endorsement of the Navy Department.

The items of the battery are also known as tests of postural equilibrium-disequilibrium (Graybiel & Fregly 1966).

All had undergone a comprehensive medical and otological evaluation. The very great majority also had benefit of a comprehensive vestibular evaluation.

Table I Correlation of ataxia test battery scores with chronological ages of normal men

Ataxia test battery	Age categories											
	16-60		16-30		31-40		41-45		46-50		51-60	
	N	r	N	r	N	r	N	r	N	r	N	r
SR	1 055	0.97 ^a	547	0.91	100	0.76	125	0.41	241	0.44	42	0.24
Walk E/O	1 055	0.90 ^a	547	0.87	100	0.72	125	0.78	241	0.47	42	0.85
Stand E/O	1 055	0.66 ^a	547	0.58	100	0.86	125	0.80	241	0.18	42	0.14
Stand E/C	1 055	0.63 ^a	547	0.98	100	0.49	125	0.68	241	0.82	42	0.88
SOLEC-R	749	0.13	488	0.61	85	0.26	36	0.87	105	0.29	35	0.68
SOLEC-L	749	0.08 ^a	488	0.39	85	0.61	36	0.98	105	0.00	35	0.59

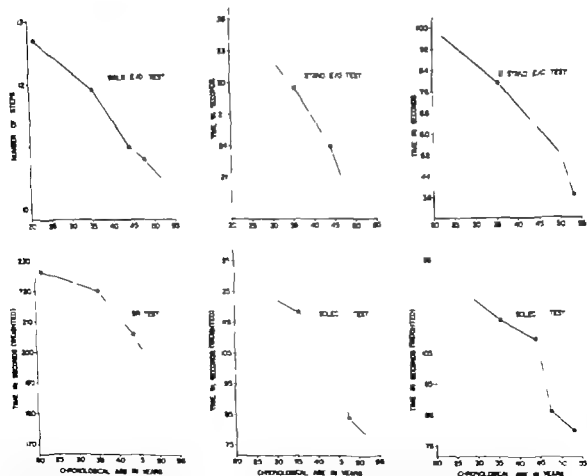
 $p < 0.001$ 

Fig 1 Chronological aging influences on ataxia test battery performance scores. (A) Walking heel-to-toe with eyes open on a 3/4-inch-wide, 8-foot-long rail. (B) Standing heel-to-toe with eyes open on the 3/4-inch-wide rail. (C) Standing heel-to-toe with eyes closed on a 2 1/4-inch-wide, 30-inch-long rail. (D) Sharpened Romberg: standing heel-to-toe on the floor with eyes closed. (E-F) Stand One Leg Eyes Closed-Right and -Left on the floor

Table II *Walk E/O test Norms, means and standard deviations of scores of normal men in five age categories*

Percentile	Age categories				
	16-30	31-40	41-45	46-50	51-60
99th	15	15	15	15	15
95th	—	—	—	—	—
90th	—	—	—	—	14
85th	—	—	14	14	—
80th	—	—	13	13	13
75th	—	14	—	—	—
70th	—	—	—	—	12
65th	14	—	12	—	11
60th	—	13	—	1	—
55th	—	—	—	—	10
50th	13	—	11	11	—
45th	—	12	—	—	—
40th	—	—	10	10	—
35th	12	11	—	—	9
30th	—	10	—	9	—
25th	11	—	9	—	8
20th	—	—	—	8	—
15th	10	9	8	7	7
10th	9	7	7	—	—
5th	8	6	6	5	—
4th	7	—	—	—	—
3rd	—	—	—	—	—
2nd	—	4	3	4	4
1st	6	—	—	—	—
Age, N	547	100	135	41	42
Mean	21.43	35.52	44.00	47.65	52.98
S.D.	3.4	3.01	1.12	1.36	2.01
Score (maximum 15 steps)					
Mean	12.69	11.89	11.00	10.83	10.38
S.D.	2.42	2.82	2.66	2.99	2.71

Table III *Stand E/O test Norms, means, and standard deviations of scores of normal men in five age categories*

Percentile	Age categories				
	16-30	31-40	41-45	46-50	51-60
99th	166	173	107	76	41
98th	148	143	100	68	39
97th	130	113	79	55	38
96th	118	93	73	49	37
95th	105	78	67	43	36
90th	72	63	45	35	28
85th	53	48	37	27	26
80th	47	37	32	4	25
75th	4	3	29	2	23
70th	38	29	26	1	22
65th	34	27	24	20	21
60th	31	5	2	19	20
55th	29	3	20	18	18
50th	27	21	18	17	18
45th	26	19	17	16	17
40th	24	18	16	15	16
35th	22	17	14	14	15
30th	20	16	13	13	13
25th	19	15	1	12	12
20th	17	13	11	11	11
15th	16	12	10	—	10
10th	14	10	9	10	9
5th	1	9	8	9	8
4th	11	8	—	—	—
3rd	10	—	7	8	—
2nd	9	6	5	—	7
1st	—	5	4	—	5
Age, N	547	100	135	41	42
Mean	21.43	35.52	44.00	47.65	52.98
S.D.	3.4	3.01	1.12	1.36	2.01
Score (maximum 180 seconds)					
Mean	36.33	29.50	3.88	19.90	18.12
S.D.	30.24	28.73	19.47	14.66	7.99

military and civilian scientific, medical engineering aviation and technical personnel.

Method

Because detailed administration and scoring procedures have been published previously (Fregly in press, Fregly & Graybiel, 1968; Fregly et al., 1971; Graybiel & Fregly 1966) these tests are described only briefly below. All tests were undertaken while subjects wore hard-soled shoes and were in the stringent position of arms folded against chest, feet aligned heel-to-toe (tandemly), with the exception of the Stand One Leg Eyes Closed tests, and body

erect or nearly erect. Administered in the following order they consisted of the

1. Sharpened Romberg (SR) standing on floor with eyes closed for 60 seconds. Minimum score obtainable, 240 seconds.
2. Walk Eyes Open (Walk E/O) walking steps per trial on a 3/4-inch-wide by 8-ft long rail. Maximum score obtainable: steps.
3. Stand Eyes Open (Stand E/O) standing the 3/4-inch-wide rail for a maximum 60 seconds per trial. Maximum score obtainable 180 seconds.
4. Stand Eyes Closed (Stand E/C) standing

Table IV Stand E/C test Norms, means, and standard deviations of scores of normal men in five age categories

Percentile	Age categories				
	16-30	31-40	41-45	46-50	51-60
99th	180	180	180	180	121
98th	—	—	—	—	119
97th	—	—	—	—	102
96th	—	—	179	179	101
95th	—	—	—	165	100
90th	—	179	152	135	82
85th	178	163	131	91	75
80th	163	148	120	75	51
75th	150	127	97	65	44
70th	140	118	72	54	37
65th	127	100	62	48	35
60th	115	78	56	43	33
55th	106	63	47	39	31
50th	97	56	43	34	30
45th	78	49	37	29	28
40th	70	43	34	28	26
35th	62	36	30	25	25
30th	55	32	27	23	23
25th	48	29	24	21	21
20th	43	26	21	20	19
15th	35	23	19	18	17
10th	30	20	17	16	14
9th	22	16	13	14	11
4th	20	15	12	13	10
3rd	18	14	11	—	—
2nd	16	—	10	11	9
1st	14	—	9	10	—
Age, N	547	100	125	241	42
Mean	21.43	35.52	44.00	47.65	57.98
S.D.	3.24	3.01	1.12	1.36	2.01
Score (maximum 180 seconds)					
Mean	98.01	79.29	63.57	51.37	37.50
S.D.	55.32	58.03	51.89	44.84	26.86

Table V SR test Norms, means, and standard deviations of scores of normal men in five age categories

Percentile	Age categories				
	16-30	31-40	41-45	46-50	51-60
99th	240	240	240	240	40
95th	—	—	—	—	—
90th	—	—	—	—	—
85th	—	—	—	—	—
80th	—	—	—	—	—
75th	—	—	—	—	—
70th	—	—	—	—	239
65th	—	—	—	—	—
60th	—	—	—	39	238
55th	—	—	—	—	223
50th	—	39	239	238	208
45th	—	—	—	211	198
40th	239	—	238	195	192
35th	—	238	—	187	186
30th	—	—	207	183	146
25th	38	221	191	157	134
20th	228	212	183	143	101
15th	202	205	135	106	76
10th	189	158	92	68	69
9th	161	105	64	39	28
4th	145	95	58	31	27
3rd	134	83	42	26	26
2nd	96	80	35	23	25
1st	55	50	29	17	4
Age, N	547	100	125	241	42
Mean	21.43	35.52	44.00	47.65	52.98
S.D.	3.24	3.01	1.1	1.36	2.01
Score (maximum 40 seconds)					
Mean	226.70	220.33	205.95	190.29	181.00
S.D.	33.95	42.57	59.77	67.36	71.62

a 2 1/4-inch-wide by 30-inch-long rail for a maximum of 60 seconds per trial. Maximum score obtainable 180 seconds.

- 5 and 6. Stand One Leg Eyes Closed (SOLEC-R and SOLEC-L): standing stationary on the floor on each leg for a maximum of 30 seconds on any trial. Maximum score obtainable 150 seconds on each leg.
7. Walk On Floor Eyes Closed (WOFEC): walking in the stringent position described above for a maximum of 10 steps per trial. Maximum score obtainable 30 steps.

This battery of ataxia tests evolved in stages. Whereas all subjects undertook the rail battery and the SR test, as noted above, a small

number also undertook the SOLEC tests (N 749) and fewer still also undertook the WOFEC (N 287).

RESULTS

Chronological age influences on ataxia test battery scores

To control for the negative influences of chronological aging on performance it was necessary to demonstrate a nonsignificant correlation between chronological age and scores on each of the test battery items within each of several age groupings. Five age groups were found to be required to accomplish this end, as shown in Table I. The mean ataxia test

Table VI. SOLEC R test Norms, means, and standard deviations of scores of normal men in five age categories

Percentile	Age categories				
	16-30	31-40	41-45	46-50	51-60
99th	150	150	150	150	150
95th	—	—	—	—	—
90th	—	—	—	149	149
85th	—	—	—	149	131
80th	—	—	—	138	128
75th	—	—	—	127	114
70th	—	—	149	124	111
65th	—	—	—	117	102
60th	149	149	148	105	90
55th	—	148	145	87	79
50th	148	147	135	77	69
45th	146	140	128	71	59
40th	141	128	115	65	55
35th	133	112	106	49	49
30th	129	100	72	43	44
25th	124	91	65	37	40
20th	115	78	56	34	33
15th	95	62	45	30	29
10th	75	37	39	27	24
5th	54	31	22	23	17
4th	50	30	20	22	16
3rd	44	29	18	20	—
2nd	39	—	17	18	15
1st	34	27	15	11	14
Age, N	488	85	36	105	35
Mean	21.25	35.71	43.75	47.83	53.14
S.D.	3.10	2.95	1.42	1.43	2.11
Score (maximum 150 seconds)					
Mean	129.87	118.39	110.53	83.91	76.77
S.D.	31.68	4.02	46.61	46.93	43.61

Table VII. SOLEC L test Norms, means and standard deviations of scores of normal men in five age categories

Percentile	Age categories				
	16-30	31-40	41-45	46-50	51-60
99th	150	150	150	150	150
95th	—	—	—	—	—
90th	—	—	—	—	149
85th	—	—	—	149	137
80th	—	—	—	140	128
75th	—	—	—	129	123
70th	—	149	149	125	110
65th	—	148	148	118	101
60th	—	145	146	110	85
55th	—	139	141	97	74
50th	149	130	138	84	70
45th	148	127	133	71	67
40th	142	123	125	57	60
35th	133	114	101	52	56
30th	127	100	81	48	45
25th	122	91	65	4	41
20th	113	68	53	38	38
15th	98	60	41	34	34
10th	78	44	22	27	29
5th	56	37	19	22	17
4th	53	35	—	20	16
3rd	42	34	18	18	—
2nd	31	31	12	16	15
1st	24	22	10	13	14
Age, N	488	85	36	105	35
Mean	21.25	35.71	43.75	47.83	53.14
S.D.	3.10	2.95	1.42	1.43	2.11
Score (maximum 150 seconds)					
Mean	130.12	115.55	109.26	86.17	80.26
S.D.	31.70	40.23	48.86	46.69	43.51

scores within each new chronological grouping are plotted in Fig. 1.² It may be seen in Table I that in the total group of subjects tested (16-60 year olds) all correlations between chronological age and subtest scores were highly significant, whereas none of the consistently lower correlations within the five subgroups were statistically significant.

Revised ataxia test battery normative standards

Mean ataxia test battery subtest scores and standard deviations, and the raw scores and

WOFEC scores by ages are not included because of their very low and not statistically significant correlation (r 0.06) with age (17-61 year old men) (Fregly et al., 1971).

their percentile equivalents for each of the five new chronological age groupings are shown in Tables II to VII. Included in each table are the mean ages and standard deviations. For the convenience of users of the ataxia test battery the recently published WOFEC test norms (Fregly et al., 1971) are reproduced in form similar to the above in Table VIII. Also for convenience sake, all scores arbitrarily defined as "ataxic" (Fregly & Graybiel 1970; Fregly et al. 1971) i.e., all 5th percentile and below cut-off scores (4th percentile and below for WOFEC) on each ataxia test battery item for each of the five chronological age groupings, are set off by a line between the 10th and 5th percentile in Tables II to VII. It should be emphasized, as

Table VIII. WOFE \bar{C} test. Norms mean, and standard deviation of scores of normal men, ages 17-61 years

Percentile	Score
99th	30
95th	—
90th	—
85th	—
80th	—
75th	—
70th	—
65th	—
60th	—
55th	—
50th	—
45th	—
40th	—
35th	—
30th	—
25th	—
20th	—
15th	—
10th	—
5th	—
4th	29
3rd	27
2nd	24
1st	17
Age, N	287
Mean	24.25
S.D.	8.73
Score (maximum 30 steps)	
Mean	29.74
S.D.	1.63

reported previously (Fregly & Graybiel, 1970; Fregly et al., 1971) that, whereas a raw score having a 5th percentile, or below equivalent has been defined as an ataxic test score, an individual is considered by us to be frankly ataxic (Fregly et al., 1971) only when scores on all items of the battery performed with eyes

A perfect score of 30 steps was obtained by 96% of the standardization group and, therefore, has a 100th-5th percentile equivalent (Fregly et al., 1971).

This refers to retesting an individual to insure the reliability of 5th percentile or below scores obtained during initial testing.

A determination of expected medical and other health-related correlates of aging by data analysis of carefully evaluated variables would undoubtedly throw desirable light on the nature of the observed group differences in terms other than aging per se; e.g. in terms of physiological aging. Preliminary efforts in this direction have begun (Fregly et al., 1968; Fregly et al., 1972).

closed (Stand E/C, SR, SOLEC R and SOLEC L, and WOFE \bar{C}) fall at or below the 5th percentile level (4th percentile for WOFE \bar{C} indicated by dotted line in Table VIII,¹ and then only after "testing the limits"² (Fregly et al., 1971).

COMMENT

Previously published normative standards of performance on all subtests of the "rail battery" (Graybiel & Fregly 1966) and on all subtests (except WOFE \bar{C}) of the "floor battery" (Fregly & Graybiel, 1968) were based on the following chronological age groupings, 17-42, 43-50 and 51-53 years. The new chronological age groupings on which the revised normative standards are based tentatively suggest that aging³ negatively influences ataxia test battery performance skills several years earlier than previously reported (Fregly & Graybiel, 1968; Graybiel & Fregly 1966): i.e., within the 30-40 rather than 43-50 year age range.

Noteworthy is the finding that amidst great individual differences in the performance scores of the men tested (Tables II to VII as well as amidst differences in the nature of the complex performance skills sampled by the varied subtests, the magnitude of the negative influences of aging observed on all subtests (except WOFE \bar{C}) performance scores was very nearly similar (Fig. 1). This result implies a reliably unitary sensitivity of the ataxia test battery to aging influences on the complex postural equilibrium functions sampled by it. However the ultimate reliability of all chronological age group differences in performance scores on this ataxia battery awaits testing of additional samples of comparable individuals (particularly in the 51-60 age group).

A revision of normative standards of ataxia test battery performance scores of samples of women is needed also, particularly for clinical purposes, and awaits availability of larger samples of testees who are comparable to the samples of men tested in terms of medical and physical status. Meanwhile, present revised

normative standards based on new chronological age groupings of the maximum number of normal men tested to date at this laboratory which are now more nearly similar to the chronological age groupings employed for women—ages 18–29, 30–49 and 50–59 (Fregly & Graybiel 1968, Graybiel & Fregly 1966)—should allow greater comparability of the scores of men with those of women, as well as generally more precise application of the test battery in both laboratory and clinical situations. It has proved to be particularly useful in vestibular physiological and neuro-otological assessment of postural equilibrium-disequilibrium including spontaneous and induced vestibular ataxia (Fregly in press, Fregly & Graybiel, 1968, Fregly & Graybiel, 1970, Fregly et al. 1971, Graybiel & Fregly 1966). Other clinical and/or research applications of the battery such as general medical, physiological, pharmacological, gerontological, audiological, and psychological are recommended.

ACKNOWLEDGMENT

The technical assistance of Mr Theron L. Trimble and the special assistance of Dr Herman J. Schaefer are gratefully acknowledged.

ZUSAMMENFASSUNG

Revidierte normative Leistungsziffern männlicher Versuchspersonen im Alter von 16–60 Jahren werden hier vorgelegt, die sich auf eine quantitative Ataxie Testserie an 1035 normalen Versuchspersonen aufteilen in fünf neue Altersgruppen beziehen. In allen Teiltesten der Serie wurde eine sehr ähnliche Empfindlichkeit für Altersinflüsse beobachtet, welche eine gleichförmige Reaktion der komplexen Gleichgewichts-

funktion der Körperhaltung auf solche Einflüsse belegt. Die Ergebnisse scheinen anzudeuten, dass Progressivität des gefundenen negativen Altersseffektes im Leben früher erscheint als bisher angenommen. Die neuen normativen Ziffern erlauben eine präzise Anwendung der Testserie in Klinik und Labor erben. Die Serie hat sich für die vestibulär-physiologische und neuro-otologische Beurteilung des Gleich- und Nichtgleichgewichts der Körperhaltung einschließlich der spontanen und induzierten vestibulären Ataxie als besonders nützlich erwiesen. Die Testserie hat sich auch andere Verwendungsmöglichkeiten im klinischen Bereich und in der Forschung.

REFERENCES

- Fregly A. R. 1968. An ataxia test battery not requiring falls. *Aerospace Med* 39, 277.
 — Vestibular ataxia and its measurement in man. In *Handbook of sensory physiology* Vol. 1, Vestibular system (ed. H. H. Kornhuber). Springer Verlag, Berlin, Heidelberg, New York.
 Fregly A. R. & Graybiel, A. 1970. Labyrinthine defects as shown by ataxia and caloric tests. *Acta Otolaryng* (Stockh.) 69, 216.
 Fregly A. R., Graybiel A. & Smith, M. J. 1972. With floor eyes closed (WOFECE): A new addition to ataxia test battery. *Aerospace Med* 43, 393.
 Fregly A. R., Mitchell R. E., Smith, M. J., Oberman A., Greene, J. W. & Graybiel, A. 1972. Correlates of postural equilibrium functions in "Thousand Aviators" Long-term follow-up. *NAAMRL Rep.* Naval Aerospace Medical Research Laboratory Pensacola, Fla., in preparation.
 Fregly A. R., Oberman, A., Graybiel, A. & Mitchell R. E. 1968. Thousand aviator study: Neurovestibular contributions to postural equilibrium function. *Aerospace Med* 39, 33.
 Graybiel, A. & Fregly A. R. 1966. A new quantitative ataxia test battery. *Acta Otolaryng* (Stockh.) 61, 292.

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INTERACTION BETWEEN THE UTRICLES AND THE HORIZONTAL SEMICIRCULAR CANALS

I. Unilateral Selective Sectioning of the Horizontal Ampullar Nerve followed by Tilting around the Longitudinal Axis

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(Received January 20, 1972)

Abstract Selective unilateral sectioning of the horizontal ampullar nerve was performed in 48 cats. This resulted in horizontal nystagmus in a contralateral direction. The animals were then tilted around their longitudinal axis, and the effect on nystagmus was studied by visual inspection and electronystagmography. If the cat was tilted to the operated side the nystagmus increased in frequency whereas if it was tilted to the opposite side the nystagmus frequency decreased. The cause of this is discussed.

In studying the function of the vestibular organ, the two systems, the semicircular canals and otolith organs, have usually been investigated separately. The semicircular canals are known to react to angular acceleration and also to release nystagmus, whereas the otolith organs are influenced by linear acceleration (Löwenstein, 1956; Taft & McNally 1934). Opinions differ however regarding the effect which the otolith organs have on the eye movements. Some authors consider that they cause only eye deviations (Versteegh, 1927; Ulrich, 1934; Szentágothai, 1952; Jongkees, 1950) other authors maintain that they can also release nystagmus (Fernández et al., 1959; Owada et al. 1960). This problem is intimately connected with another phenomenon, frequently observed in otolaryngology namely positional nystagmus (Bárány 1921). The problem remained unsolved, however owing to

the fact that the more precise influence of the otolith organs on the different eye muscles was still not known a few years ago. Fluor & Mellström (1970) have studied these problems in the cat, and have shown, with regard to the horizontal eye movements, with which this paper deals, that on the lateral part of the utricle there is an area which, during electrical stimulation, causes a horizontal eye deviation to the stimulated side. In some of the cases, however nystagmus towards the same side was also released. Consequently the results are somewhat contradictory and we do not consider the problem concerning the horizontal eye movement representation on the utricular surface as definitely solved. The question is now whether the nystagmus may depend on the fact that the stimulation current extended to the horizontal canal or that it was elicited from the utricle itself. We have therefore with another stimulation technique studied in a series of experiments, whether there is some interplay between the horizontal canal and the utricle. This paper describes how a destruction nystagmus, after unilateral selective sectioning of the horizontal ampullar nerves, is influenced by a change in the utricular activity caused by tilting along the longitudinal axis of the animal.

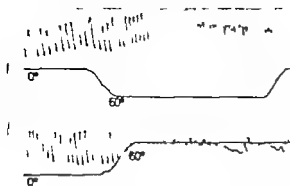


Fig. 1 Consecutive curves from a cat after selective unilateral sectioning of the left horizontal ampullar nerve, followed by tilting. The upper curve shows the electronystagmography and the lower curve shows the tilting of the table. Tilting to the right upwards, to the left downwards.

MATERIAL AND METHODS

For the experiments 48 adult cats were used. They were anesthetized with ether with a so-called Ayer system, which is employed in pediatric anesthesia. The animals were placed on a tilting table which can be tilted around different axes.

A 3 cm long vertical incision was made immediately anterior to the external ear. The external meatus was dissected free and cut close to the temporal bone, where after it was attached to the side by a self-retaining forceps. The middle ear was explored and the malleus, incus and the tensor tympani muscle were removed. The facial canal was opened and part of the nerve resected. At the bottom of the canal two small bluish spots were visible, marking the ampullae of the horizontal and the anterior vertical canals. Medial to these a fenestration was performed large enough for the visualization of the nerves from the two ampullae. The horizontal ampullar nerve was finally cut as far away from the ampulla as possible. Great care must be taken not to damage the membranous labyrinth or the ampullae, or the utricle behind. However, under the microscope it was not so difficult, since it was possible to see all the other struc-

tures and their borders and afterwards to check by visual inspection.

The recording of the eye movements was done partly by direct visual ocular inspection of the cat's eyes, and partly by traditional electronystagmography with needle electrode in the lateral canthi bilaterally and with the ground electrode in the skin of the neck.

Stimulation was applied by tilting the animals 60° around their longitudinal axis on either side.

RESULTS

In order to avoid all conceivable artefacts, the operation was performed in stages. Five cats were awakened after a short period of anesthesia without any operation being performed. The cat was then again anesthetized, and the middle ear explored. The animals were brought to a level of superficial anesthesia. Thereafter deep anesthesia was again induced and fenestration was performed but the horizontal ampullar nerve was not divided. Nystagmus did not develop in the cats at any of these stages when the animals were brought to a superficial level of anesthesia. Finally the horizontal ampullar nerve was selectively sectioned, and horizontal destruction nystagmus occurred towards the opposite side when the cats were brought to a level of superficial anesthesia.

In 16 cats tilting toward the operated side resulted in an evident increase in the frequency of nystagmus amounting to 1-10 beats/10 sec. Furthermore, where the beats had previously been somewhat irregular in shape, they now became much more regular in both frequency and amplitude. On tilting toward the nonoperated side, 14 showed a decrease in nystagmus frequency of 1-8 beats/10 sec, and in 4 cats there was immediate total inhibition of nystagmus. Eight cats showed no substantial alteration in the character of the nystagmus during tilting toward the operated side, and 10 cats when tilted toward the nonoperated side. Twelve cats showed an "ideal" reaction when

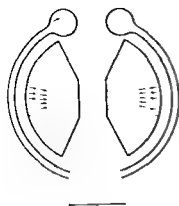


Fig. 2.

Fig. 2. Schematic picture of the utricles and horizontal semicircular canals in resting position. The arrows indicate the orientation of the haircells, i.e., the direction in which they increase their discharge frequency.

Fig. 3. Schematic picture of the utricles and horizontal semicircular canals, after unilateral selective sectioning of the left horizontal ampullar nerve, and when the animal is tilted to the left. Arrow indica-

Fig. 3.

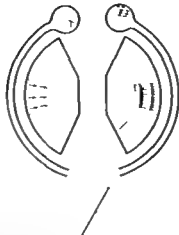


Fig. 4.

tion same as in Fig. 2. The number of arrows symbolizes discharge frequency.

Fig. 4. Schematic picture of the utricles and horizontal semicircular canals, after unilateral selective sectioning of the left horizontal ampullar nerve, and when the animal is tilted to the right. Arrow indication same as in Fig. 2. The number of arrows symbolizes discharge frequency.

tilted in either direction i.e. facilitation during tilting toward the operated ear and inhibition toward the nonoperated side (Fig. 1).

DISCUSSION

The aim of this investigation was to create a system with a functioning reflex arc only from one horizontal semicircular canal, whose activity could then be so modulated by tilting the animal that the utricular activity increased on one side and simultaneously decreased on the other. The investigation has shown that tilting changes the nystagmus by either increasing or decreasing it, depending on the position. Fluor & Mellström (1970) have shown that horizontal eye movements from the otolith organs can be induced only by stimulation of a small utricular area situated in the middle of its lateral section (Fig. 2). It must be this area which influences the activity of the canal or the reflex arc, and causes the character of the nystagmus to change.

With the aid of a few figures we shall try

to explain the course of events in the different situations. Fig. 2 shows the two semicircular canals and utriculi, and the direction in which the haircells in the two systems are oriented, i.e. the direction in which they bend when the activity increases. This is the resting position. If the cat is now tilted to the operated side, the activity is inhibited in the utricular area on that side, whereas, on the other side it is simultaneously increased. (Fig. 3). The effect on nystagmus may be due to the fact that the intact semicircular reflex arc is facilitated either by increased activity in the ipsilateral utricle or by inhibition from the contralateral utricle. This investigation does not enable us to determine which of these alternatives is operative.

If on the other hand, the animal is tilted to the nonoperated side the result is quite the opposite (Fig. 4). Here, we have an increased activity in the utricle on the operated side, and inhibition on the other. As the investigation has shown, the effect on nystagmus is also the opposite.

Consequently the investigation has raised two new questions. Are the results actually due to the utricular influence, as has been assumed in the discussion and in that case does the reaction originate from the ipsilateral or the contralateral utricle. These questions will be fully dealt with in a later paper.

ZUSAMMENFASSUNG

Bei 48 Katzen wurde der horizontale Ampullarnerv unilateral abgeschnitten. Dies resultierte in einem horizontalen Nystagmus nach die kontralateralen Seite. Danach wurden die Katzen um die longitudinale Achse gedreht, und die Wirkung auf den Nystagmus wurde mit visueller Inspektion und Elektronystagmographie studiert. Wurden die Katzen nach der operierten Seite gekippt, erhöhte sich die Geschwindigkeit des Nystagmus wenn sie nach der anderen Seite gedreht wurden, stellte man eine Verminderung der Nystagmusgeschwindigkeit fest. Die Ursache dazu wurde diskutiert.

REFERENCES

- Bárány R. 1921 Diagnose von Krankheitserscheinungen im Bereiche des Otolithenapparates. *Acta Otolaryng* (Stockh.) 2 434
- Fernández, C., Alzate, R. & Lindsay J. R. 1959 Experimental observations on postural nystagmus in the cat. *Ann Otol* 68 816.
- Fluor E. & Mellström, A. 1970. Utricular stimulation and oculomotor reactions. *Laryngoscope* 80 170
- Jongkees, L. B. W. 1950 On the function of the sacculus. *Acta Otolaryng* (Stockh.) 38 18.
- Löwenstein, O. 1956 Peripheral mechanisms of equilibrium. *Brit Med Bull* 12 114
- Owada, K., Shimizu, S. & Kimura, K. 1960. The influence of the utricle on nystagmus. *Acta Otolaryng* (Stockh.) 52 215
- Szentágothai, J. 1952. Die Rolle der einzelnen Labyrinthrezeptoren bei der Orientierung vom Kopf und Kopf im Raum. *Alumina kiado*, Budapest
- Tak, J. & McNally W. J. 1934 Some features of action of the utricular maculae and of associated actions of the semicircular canals of the frog. *Phil Trans B* 224 241
- Ulrich, H. 1934 Die Funktion des Otolithen geprüft durch direkte mechanische Beeinflussung des Utrikulusotolithen am lebenden Hecht. *Pflüger Arch Ges Physiol* 235 545
- Versteegh, C. 1927 Ergebnisse partieller Labyrinthexstirpation bei Kanarienvögeln. *Acta Otolaryng* (Stockh.) 11 393

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NYSTAGMUS RESPONSES DURING TRIANGULAR WAVEFORMS OF ANGULAR VELOCITY ABOUT THE Y AND Z AXES

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(Received March 18, 1972)

Abstract Nystagmus slow phase velocity was measured during rotation stimuli consisting of short (12 sec) triangular waveforms of angular velocity. In one test series the horizontal semicircular canals were stimulated and in another test series the vertical canals were stimulated. The data were fit by equations derived from the assumption that the responses depend upon a cupula-endolymph system that responds like an overdamped torsion pendulum. Mean long time constant (H/Δ) values found for the horizontal and vertical canal responses were, respectively 16 sec and 7 sec, consistent with previous findings, whereas gain constants found for the horizontal and vertical canals were, respectively 8.0 sec and 5.4 sec. Procedures for quickly extracting these parametric values from nystagmus records are described and, for practical reasons, the effects of stimulus distortion on the estimates of parameters were also considered.

In previous reports parameters of the subjective reaction to semicircular canal stimulation were estimated from responses to a series of test stimuli consisting of short triangular waveforms of angular velocity (Guedry et al., 1971 a, 1971 b; Owens & Guedry 1969). This method appears to offer some practical advantages in regard to reliability and ease of measurement of vestibular sensation. The feasibility of using similar test procedures, as outlined by Guedry et al. (1971 b) in order to

permit estimation of nystagmus response parameters is examined in this report. Both horizontal and vertical canal responses were evaluated and compared to determine whether or not the method is sufficiently sensitive to detect known differences in responses from the two sets of canals.

These procedures are based on a set of equations, derived from the torsion pendulum model which has been used extensively by van Egmond et al. (1949). The equations assume an ideal stimulus waveform, but in practice the waveform is often distorted due to performance limitations of rotary devices. Minor deviations from the ideal waveform are probably inconsequential, but the resulting errors may become significant when, for example fine discriminations between individuals are made or when measurements from several laboratories are compared. Therefore the effect of stimulus waveform distortion on parameter estimation was also examined in this report.

PROCEDURE

Subjects

Twenty US naval and marine officers served as subjects. These men ranged in age from 20 to 26 years. All had recently passed standard Navy flight physical examinations without signs of vestibular disorders.

This research was sponsored jointly by the US Army Aeromedical Research Laboratory and the Naval Aerospace Medical Research Laboratory.

Opinions or conclusions contained in this report are those of the authors. They are not to be construed as necessarily reflecting the view of, or the endorsement of the Departments of the Army or Navy.

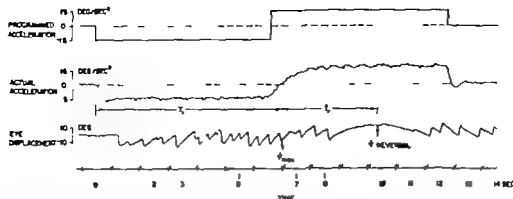


Fig 1 A sample record showing programmed acceleration, actual acceleration, and recorded eye displacement for stimulation of the vertical semicircular canals. Peak slow phase eye velocity (ϕ_{max}) is measured

approximately at the end of the acceleration period (T_0), and the time interval (t) is measured from the end of T to the point of response (ϕ) reversal.

Apparatus

The rotation device was a Stille Werner RS-3 rotator fitted with a rectangular platform and a removable chair. The device was controlled by an external command voltage generated by a low frequency waveform generator. Stimulus angular acceleration was recorded from an accelerometer mounted on the rotation platform, and angular velocity was recorded from a tachometer attached to the drive motor.

Appropriately placed Beckman electrodes were used to record corneoretinal potentials from the subjects. These potentials were led through sliprings to a Sanborn 350 recording system and displayed on a chart recorder. The time constant of the amplification system was 2.5 sec.

Stimulus angular acceleration and velocity were also recorded on the chart display and angular acceleration was recorded on magnetic tape. The angular acceleration signal was later fed into an analog computer to generate a theoretical response curve for comparison with the one obtained from the prediction equations.

Method

Responses from both the horizontal and vertical semicircular canals were tested. To stimulate the horizontal canals, the subject was seated upright in the chair so that his z-axis

was aligned with the axis of rotation. To stimulate the vertical canals, the chair was removed and the subject was positioned on his right side on the platform so that his y-axis was aligned with the axis of rotation. Half of the subjects were tested in the seated position first and half were tested in the right-side down position first.

The stimulus consisted of a triangular waveform of angular velocity formed by programming a 15 deg/sec² acceleration to last for 6 sec, followed immediately by deceleration of the same magnitude and the same duration. This stimulus was administered ten times in each position with the direction of rotation alternated from trial to trial.

Subjects were tested in total darkness with their eyes open. Mental alertness was maintained by requiring the subjects to add a series of numbers read to them during stimulation and to answer after stimulation had ceased. Eye movements were calibrated before and after each stimulus presentation. The inter trial interval was 30 sec.

The nystagmus records were scored with a device (Guedry & Turnipseed, 1967) that provides readout, in both graphical and digital form, of slow phase eye velocity as a function of time. In addition, two measurements were obtained for calculation of parameter estimates. The first measure was the magnitude

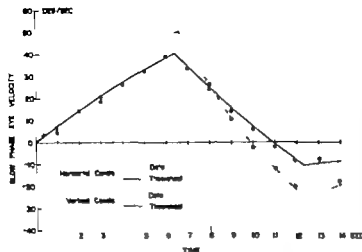


Fig 2 Nystagmus responses from stimulation of the horizontal and the vertical semicircular canals by a triangular waveform of angular velocity. Data points represent the mean of twenty subjects \times ten trials. Smooth curves represent theoretical responses calculated from the prediction equations.

of peak slow phase eye velocity (v_{\max}) and the second was the time interval (t_r) from the end of the acceleration period (T_1) to response reversal. Fig. 1 illustrates the way in which these two measures were obtained.

Calculations of parameter estimates were based on the assumption that the vestibular stimulus was a triangular waveform of angular velocity formed by a constant acceleration followed immediately by a constant deceleration of the same magnitude and duration. However the actual stimulus waveform was distorted, as shown in Fig. 1 particularly when the angular acceleration reversed direction. As a result, acceleration persisted beyond the point of initial change. Thus, for parameter estimation, the point at which acceleration passed through zero was chosen to mark the end of T_1 , the beginning of t_r , and approximately when v_{\max} should occur.

Two theoretical response curves of slow phase velocity were generated, one from the prediction equations (Guedry et al., 1971a) assuming an idealized acceleration waveform, and the other using the actual acceleration waveform recorded on magnetic tape as an input to an analog computer programmed with the appropriate second-order differential equation. The latter response curve was obtained by manipulating the values of the program to approximate as closely as possible the meas-

ured slow phase eye velocity as a function of time. Both curves were then compared in order to assess the effects of stimulus wave form distortion.

RESULTS AND DISCUSSION

Estimation of response parameters

Mean nystagmus responses for the group are shown in Fig. 2. Significant differences were found between the responses of the horizontal canals (x-axis rotation) and those of the vertical canals (y-axis rotation). The total slow phase displacement of the two responses was approximately the same, but the peak magnitudes of both acceleratory and deceleratory nystagmus were greater and the reversal occurred sooner for the vertical canal response. These differences were statistically significant ($p < 0.0005$).

According to the equations derived in previous reports (Guedry et al. 1971a, 1971b), the nystagmus response can be specified if the values of two parameters, Π/Δ and $K_n(\theta/\Delta)$, are known. The value of Π/Δ can be determined from t_r , and once Π/Δ is known, the value of $K_n(\theta/\Delta)$ can be determined from v_{\max} . In practice, it is somewhat tedious to calculate these values from the equations. Therefore nomograms were prepared, and are shown in Figs. 3 and 4. The use of these nomo-

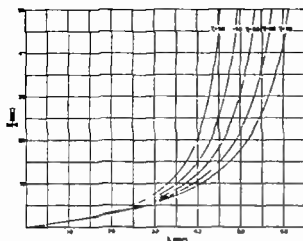


Fig. 3 Relationship between t and Π/Δ

grams is illustrated with the average values of t and ψ_{max} found in this study. For the horizontal canal response t was 4.5 sec and T_1 was 6.4 sec. Thus, the value of Π/Δ interpolated from Fig. 3 is about 16.0 sec. A $K(\theta/\Delta)$ can be estimated from Fig. 4 by locating the intersection of a 6.4 sec T_1 curve with the Π/Δ value of 16 sec to obtain the $K(\theta/\Delta)/\psi_{max}$ value of 0.2 sec²/deg. When this is multiplied by the mean ψ_{max} values of 40.2 deg/sec, a $K_s(\theta/\Delta)$ value of 8.0 sec is obtained.

For the vertical canal response, t was 3.3 sec and T_1 was 6.6 sec therefore, Π/Δ is 7.0 sec. A value of $K(\theta/\Delta)/\psi_{max}$ of 0.11 sec²/deg was then obtained from Fig. 4 which, when

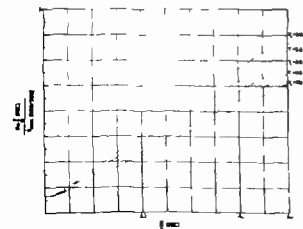


Fig. 4 Relationship between Π/Δ and $K(\theta/\Delta)/\psi_{max}$ for an angular acceleration of 15 deg/sec²

multiplied by the ψ_{max} of 49.1 deg/sec, yields $K_s(\theta/\Delta)$ of 5.4 sec.

In the same way parametric values can be obtained for other nystagmus data, once t and ψ_{max} have been measured. The nomogram were calculated for acceleration durations (T_1) of 5 to 7 sec. Pilot studies indicated that this range is of practical value when angular acceleration is 15 deg/sec² since the nystagmus response is difficult to score if the duration of acceleration is much less than 5 sec, and the errors introduced by response adaptation (Younis & Oman 1969) may become large if acceleration lasts much longer than 7 sec.

The Π/Δ determinations from Fig. 3 are independent of the magnitude of acceleration but the $K(\theta/\Delta)$ values can be obtained from Fig. 4 only for an acceleration rate of 15 deg/sec². For other acceleration rates, the correct $K(\theta/\Delta)$ value can be calculated from Fig. 4 if the value of $K(\theta/\Delta)$ found from the figure is multiplied by 15 deg/sec² and then divided by the acceleration rate actually used.

The estimates obtained here for Π/Δ are generally consistent with those found by other investigators using various methods (Benson & Guedry 1971; Guedry et al., 1971; Melville Jones et al. 1964) thus supporting the validity of this procedure. Accurate estimates of $K_s(\theta/\Delta)$ have heretofore not been readily available although calculation from data of Collins & Guedry (1967) collected during prolonged angular acceleration resulted in similar values. Further support for this procedure comes from the close fit to the data of the theoretical response curves generated by the prediction equation. Fig. 2 shows that the equation accurately predicts the horizontal canal responses and for the most part, vertical canal responses. However with vertical canal stimulation there is a fairly pronounced departure of the rise in response during the initial acceleration from the theoretical rise curve. This may be attributable to difficulties in recording and scoring vertical nystagmus (Benson & Guedry 1971; Ford, 1959; Melville Jones et al. 1964) but it may also be due to a real departure

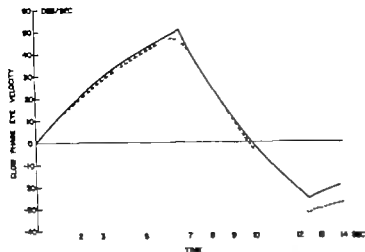


Fig. 5 Effect of stimulus waveform distortion during vertical canal stimulation. — Theoretical response curve generated by the prediction equation assuming a perfect triangular waveform stimulus; ---- Theoretical response curve generated by an analog computer from the actual stimulus used in this study

from the theoretical prediction. Nystagmus elicited by passive movement lacks the preparatory eyelid and brow movements which naturally accompany voluntary head rotations about the y-axis. The absence of these and other sensory data usually present in natural active movement may contribute to slow rise of the response, but, alternatively the fact that the nystagmus velocity waveform approximates the shape of the stimulus velocity waveform more clearly than does its theoretical counterpart may reflect a need for restructuring some theoretical concepts.

Effects of stimulus waveform distortion

It was shown in Fig. 1 that the rotation device used here does not reproduce exactly the programmed stimulus waveform. Distortion was more severe during y-axis stimulation than during z-axis stimulation due to the greater rotational inertia that occurs when the subject's body is in the right-side-down position. Any amount of stimulus distortion is undesirable nevertheless, it is a common problem with the rotary devices available in most vestibular laboratories, so an attempt was made to assess its effects on response prediction. In Fig. 5 the theoretical response curve obtained from the equation (assuming a perfect triangular stimulus waveform) is compared with the theoretical response curve generated on an analog

computer using the actual distorted stimulus waveform recorded during vertical canal (y-axis) stimulation. This amount of distortion had little effect, as indicated by the similarity of the two response curves. It introduced some errors when estimating response parameters from the equations, but these were within an acceptable margin for most purposes. Using the actual stimulus waveform, Π/Δ was estimated to be 7.3 sec and $K_n(\theta/\Delta)$ was 6.5 sec, whereas using the programmed waveform, Π/Δ was 6.8 sec and $K_n(\theta/\Delta)$ was 5.4 sec.

Individual differences

Parametric values were calculated in this study from the averaged responses of a group of subjects, but the procedure is applicable as well to the evaluation of individual responses. Large differences were found among the 20 subjects in this sample estimates of Π/Δ ranged from 9.9 to 32.7 sec for the horizontal canals and from 5.5 to 10.6 sec for the vertical canals, $K_n(\theta/\Delta)$ ranged from 4.9 to 11.2 sec for the horizontal and from 4.2 to 8.2 sec for the vertical canals.

These estimates appear to be reliable. From measures of intrasubject variability the 95% confidence interval for t was found to be ± 0.20 sec for the horizontal canals and ± 0.23 sec for the vertical canals. The 95% confidence interval for v_{max} was ± 5.68 deg/sec for the

horizontal canals and ± 8.05 deg/sec for the vertical canals. These confidence intervals were achieved from averages based on 10 trials. Greater precision could be gained by using averages over more than 10 trials, but the test procedure would be lengthened and its clinical potential usefulness diminished. Also a significant decrease in the t measure was noted with repeated presentations of the stimulus. This decrease was only 0.2 to 0.3 sec over the 10 trials, but it might have been larger and consequently more troublesome if more trials had been presented (Groen, 1957). It is unlikely that the decrease was due to a decline in alertness over trials because the other response measure, v_{max} , was not significantly affected.

The present and the preceding reports (Guedry et al. 1971a, 1971b; Owens & Guedry 1969) have described procedures for estimating parameters associated with both nystagmus and subjective responses to semicircular canal stimulation. These tests appear to be both reliable and relatively simple to administer and score.

ACKNOWLEDGMENTS

The authors wish to express their appreciation to G. Turnipseed, J. W. Norman and D. J. Gripla for their assistance in collecting and analyzing experimental data, and to Dr Margaret Smith for her assistance with statistical analysis.

ZUSAMMENFASSUNG

Parameter der Nystagmusreaktion wurden mittels eines Testverfahrens abgeschätzt, das kurze, dreieck förmige Wellenformen der Winkelgeschwindigkeit benutzte. Schätzungen des Mittelwertes wurden in folgender Weise bestimmt: $\Pi/\Delta = 16.0$ Sek und $k_{\omega}(\Delta) = 8.0$ Sek. für die horizontalen Bogengänge und $\Pi/\Delta = 7.0$ Sek. und $k_{\omega}(\Delta) = 5.4$ Sek. für die vertikalen Bogengänge. Die Π/Δ Werte stimmen mit denen anderer Methoden überein. Werte für $k_{\omega}(\Delta)$ sind bis

her nicht ermittelt worden. Bestimmung der Wirkungen von Retverzerrung auf die Werte der Reaktionsparameter und Abschätzungen der inter- und intraindividuellen Variabilität werden vorgelegt, ebenfalls Nomogramme, die eine einfache und genaue Methode für die Berechnung von Π/Δ und $k_{\omega}(\Delta)$ erlauben.

REFERENCES

- Benson, A. J. & Guedry F. E. 1971 Comparison of tracking task performance and nystagmus during sinusoidal oscillation in yaw and pitch. *Aerospace Med* 42 593.
- Collins, W. E. & Guedry F. E. 1967 Duration of angular acceleration and ocular nystagmus from cat and man. I. Responses from the lateral and the vertical canals to two stimulus durations. *Acta Otolaryng* (Stockh.) 64 373.
- Egmond, A. A. J. van, Groen, J. J. & Jongkees, L. B. W. 1949 The mechanics of the semicircular canal. *J Physiol* 110 1.
- Ford, A. 1959 Significance of terminal transients in electro-oculographic recordings. *Arch Ophthalmol* 61 899.
- Groen, J. J. 1957 Adaptation. *Pract Otorhinolaryng* (Basel) 19 54.
- Guedry F. E. & Turnipseed, G. T. 1967 Two devices for analysis of nystagmus. *Ann Otol* 77 1071.
- Guedry F. E., Stockwell, C. W. & Gilson, R. D. 1971a. Comparison of subjective responses to semicircular canal stimulation produced by rotation about different axes. *Acta Otolaryng* (Stockh.) 72 101.
- Guedry F. E., Stockwell, C. W., Norman, J. W. & Owens, G. G. 1971b The use of triangular waveforms of angular velocity in the study of vestibular function. *Acta Otolaryng* (Stockh.) 71 439.
- Melville Jones, G. Barry W. & Howalsky H. 1964 Dynamics of the semicircular canals compared in yaw, pitch, and roll. *Aerospace Med* 35 984.
- Owens, G. G. & Guedry F. E. 1969 Assessment of semicircular canal function. II Individual differences in subjective angular displacement produced by triangular waveforms of angular velocity. NAMI 1074 USAARL 69-13 Pensacola (Fla) Naval Aerospace Medical Institute.
- Young, L. R. & Oman, C. W. 1969 Model for vestibular adaptation to horizontal rotation. *Aerospace Med* 40 1076.

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MICROFILM STORING TECHNIQUE IN ELECTRONYSTAGMOGRAPHY

Two Years of Experience

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(Received April 5 1972)

Abstract. At the Regional Hospital, Örebro, Sweden, an EEG routine was established 4 years ago including ADB and integrated microfilm technique. Two years ago the microfilm technique was also applied to the ENG routine, 3 1/2 microfilm cards being used for storing records and typewritten data. Information is regained by means of a microfilm reader apparatus or a microfilm projector paper copies may also be made. The microfilm technique permits much easier and more rapid examination of previous records compared with the original paper diagram evaluation. This is important in the daily handling of the ENGs, especially when estimating possible progress or regression in the course of various vestibular diseases. The microfilm method also has reduced storing space requirements to a minimum.

The storing of various documents concerning patients has become a great problem in hospitals. The archives increase at an accelerating speed. In Sweden an official committee has presented (1968) an analysis concerning the problem of medical archives (called "Arkiv inom hälso- och sjukvård"). From this investigation a few examples may be mentioned. A 500-bed hospital is assumed to require 75 shelf metres a year for the storage of additional patient journals. The archives of an X-ray department with 50 000 investigations a year will increase by 143 shelf-metres. An EEG laboratory needs 12 shelf metres to store one year's production of 2 500 records; an ECG laboratory will fill up 25 shelf metres with 90 000 ECGs. One can only partly solve

the problem by weeding out papers. Legal rules protect the different sorts of medical documents: one has to keep them in the archives for a varying number of years, the time being dependent on the type of document and country concerned.

The committee mentioned above has not proposed any outlines concerning ENGs but it seems natural to follow the same suggestions as for EEG records keeping them in the archives for 10 years. Because of the continuously increasing problem of archive enlargement one is forced, however to consider other solutions. Such a method is microfilm technique.

In the Department of Clinical Neurophysiology of the Regional Hospital in Örebro a microfilm method was established 4 years ago, the equipment since then being used in the daily EEG routine integrated with an ADB technique. From these 4 years the experiences are very encouraging, as stated by Sörbye (1972). The method involves microfilm photographing of all relevant information written in the referral card, the total report, selected parts of the record, calibration markings and the recording schemes. All this information is kept for the future in the small film square in a data card which also contains automatically punched information, such as the patient's name and other identification data, the latter

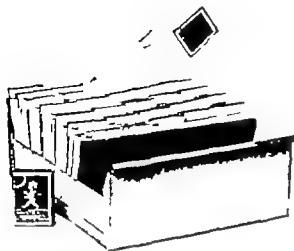


Fig. 1 The archive box, containing about 1 000 microfilm cards. One microfilm square is seen.

to be read visually or by computer. In this connection it should be mentioned that from the visually interpreted EEG record the computer is producing an automatically written report as well as storing the patient and EEG data on magnetic tape, the latter affording the possibility of later statistical investigations, etc. Concerning ENG records, the general handling of these up to now has varied from place to place. In some hospitals the records are thrown away after the report has been written, in other clinics they are kept for some time. Until 2 years ago the records of the Otoneurological Laboratory of the ENT Clinic in Örebro were not archived due to lack of archive space. It was obvious, however, that the information of the records could be valuable even after long time. Therefore it was considered a good idea to try the microfilm technique which had already shown its great value in the EEG routine. With this method the main advantage would be the space saved in the archives. The margin of this area of the Otoneurological Laboratory was so small that even the actual production of 700 ENG records a year certainly was of importance. After microfilm photography the total produc-

tion of 3 years can be stored in an ordinary shoe box (Fig. 1).

METHOD

During the past 2 years the following ENG microfilm routine has been used. A short case history and the result of the ENG investigation are photographed together with all the relevant parts of the records. The dry and fully mounted microfilm card comes out 45 seconds after a button being pushed on the microfilm camera, the next record in the meantime being prepared for photography. The identification data, etc. are written at the top of the film card with an ordinary typewriter, the card then being placed in the archive after a routine control of the data and the quality of the reproduction.

The microfilm camera of the EEG laboratory has been used for photographing (3 M 2 000 from 3 M Company).

For retrieval of information from the microfilm card there are several possibilities:

1. Microfilm reader apparatus

The enlarged film square should be visible in total on the screen, to get the best survey of the records and the best basis for estimating asymmetries, etc. This is the case in the Normandale model 1518 (WST) where the screen measures 36×43 cm (0.16 m²) meaning a fourfold reduction of the original area which has been photographed. Another apparatus of the same type is "Caps" from Ingut Company.

2. Paper copies

These can be produced in a reader-printer for instance Filmac 200 (3 M Company), giving an area of 61×45 cm (0.27 m²), which means about half the original area.

3. Microfilm projector

A projector specially constructed for microfilm cards gives good sharpness up to the picture size of at least 4×6 m. This opens up

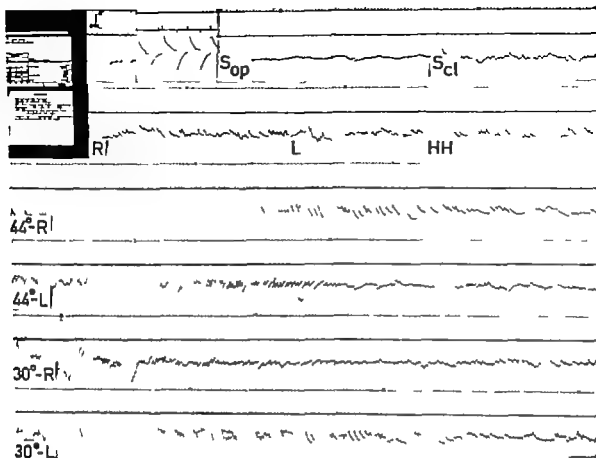


Fig 2 Enlarged microfilm squares. ENG With closed eyes (S) spontaneous nystagmus to the right is seen. In the right, left and hanging head positions (R, L and HH) there is also a nystagmus to the right. In

the caloric tests a slight symmetrical preponderance to the right is observed. (Woman, 41 with a 6 month history of rotary vertigo attacks)

possibilities for demonstration of ENG records even for larger groups.

Using the greatest reducing factor of the camera ($24\times$) the area at disposal for photography is 91×68 cm, i.e. 0.62 m². A small part of this area is used for the patient identification data, the concentrated case history and a short report of the result of the ENG investigation (typewritten so far). Disregarding the upper left corner containing this information (Fig. 2) the area to be occupied by the ENG record still exceeds 0.5 m². In this area a more than 5 metres strip of paper measuring 12 cm across may be photographed. With such a paper from an Elema Schönander "Mingograph 81" a 2-channel registration of horizontal nystagmus will be suitable: one with short

and one with long time constant. Moreover one may add 2 further channels, for instance for vertical nystagmus and vestibulospinal reflex.

We want to stress the importance of marking the different parts of the ENG carefully before microphotography. This can be done by means of large rubber stamps for each relevant part of the record (concerning the marking letters, see within parentheses after the different head positions stated below).

As shown in Fig. 2, a representative strip of paper has as a rule been microphotographed from each of the following registrations (when nothing else is stated the patient is lying supine with a 30 degree flexion of the neck. S):

1 Calibration. With eyes starting from zero position a 5 degree "horizontal" deviation to each side has on the paper as a rule been given a total amplitude of 10 mm (CAL).

2 Patient looking at a point 30 degrees "horizontally" deviating from the zero position, right and left alternatively

3 Patient fixating a point on the ceiling at zero position (S, op)

4 Same, but eyes closed (S, cl)

5 Head turned to the right, eyes closed (R).

6 Head turned to the left eyes closed (L)

7 Head hanging down eyes closed (HH).

8-11 Four registrations of the caloric reactions from stimulating right and left ear with water of 44 and 30 degrees (44 —R, etc.) stimulation time being 40 sec with a constant water flow (Oto-term from Kifa)

In the microfilm square, parts 1-7 of the registration will take two of the six possible horizontal strips, the last four being left for the caloric tests, one for each of parts 8-11. When photographing these 4 registrations their corresponding points of water flow ending should be placed on the same vertical line. Hereby one has the great advantage of easy comparison of the results concerning duration, amplitude, frequency and speed of the slow phase. This is important the main interpretation of the caloric reactions being based on a comparison between these single parameters.

DISCUSSION

In many vestibular diseases, e.g. vestibular neuronitis, streptomycin and dihydrostreptomycin lesions, Ménière's disease and suspected acoustic neurinomas, it is important following up a series of ENG's over a long period of time. For interpreting possible progress or regression in a certain patient it is certainly valuable to be able to examine the previous records themselves and not only the descriptions. Also when consecutive reports record a shift in the direction of nystagmus it is valuable to have the records kept for control concerning possible mistakes.

Microfilm technique not only offers the possibility of keeping a large number of records in a small archive unit, it also permits examination of the registrations in a shorter time and with a better total view of eventual changes than before. The unfolding and arranging of previous ENG papers is no longer necessary. Paper copies of the microfilm square may follow the patient when being referred to another clinic. Such copies may also be added to the report to the referring colleagues. This has proved to increase their interest in the investigation and to widen their understanding of the interpretation. Projection permits easier demonstration for larger groups, especially when discussing details in a series of investigations.

The method described represents new possibilities for the handling of ENG information in an easier way. At the same time the archive problems are solved. In short, the microfilm technique has proved to save space, time and trouble.

RÉSUMÉ

À l'Hôpital Régional d'Örebro (Suède) on fait depuis quatre ans des EEG de routine. On en tire un microfilm et confie à un ordinateur (ADB) les enregistrements. Depuis deux ans le microfilm a été utilisé de la même façon pour les Electro-oculogrammes. On a employé des cartes de microfilm (3 M) pour les diagrammes et les informations écrites. L'examen du microfilm peut être fait au moyen d'un lecteur ou d'un projecteur de microfilms — ou on peut en faire une photocopie. L'examen d'un ENG ainsi traité prend moins de temps que la lecture d'un document original et la comparaison entre plusieurs ENG est aussi plus facile à faire, ce qui est important, puis qu'on a souvent besoin de comparer une série d'ENG pratiqués à différents stades de la maladie pour estimer l'évolution de différents troubles vestibulaires. Le microfilm a aussi l'avantage de réduire l'espace de rangement au minimum.

ZUSAMMENFASSUNG

Seit vier Jahren werden im Landeskrankenhaus in Örebro, Schweden, die Elektroencephalogramme routinemäßig auf Mikrofilm gelagert und die Resultate durch integriertes ADB-Verfahren geliefert. Seit zwei Jahren haben wir die Mikrofilmmethode auch für Elektronystagogramme (ENG) verwendet. Es werden 3 M Mikrofilmkarten für die Lagerung der Diagramme

und maschinengeschriebene Daten verwendet und zum Lesen dieser Karten Mikrofilm-Leseapparat, Mikrofilm-Projektor oder auch Papierabzüge. Die Mikrofilmmethode erlaubt behändigere und schnellere Untersuchung früherer Aufnahmen im Vergleich mit dem Studium von Originaldiagrammen, was von Bedeutung in der täglichen Arbeit mit ENG ist, wo man oft eine Folge von Diagrammen untersuchen will um einen eventuellen Progreß oder Regreß im Verlauf von Krankheiten im Vestibularsystem aufzudecken. Die Mikrofilmmethode hat den Bedarf an Lagerraum auf ein Minimum herabgebracht und ermöglicht Demonstration von Diagrammen an der Wand mit einem Projektor.

REFERENCES

- Statens offentliga utredning 1968. *Arkiv boken hälso- och sjukård*
 Sörbye, R. 1972. In press.
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TEMPORAL INTEGRATION OF ACOUSTIC ENERGY IN PATIENTS WITH PRESBYACUSIS

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(Received April 6 1972)

Abstract The ability to integrate acoustic energy over time, i.e. temporal integration, can be measured by short tone audiometry. Abnormal temporal integration has been considered as one of the reasons for reduced discrimination in hearing impaired ears. The results of short tone audiometry in 46 persons with presbycusis are reported. The frequencies 500 Hz, 1 000 Hz, 4 000 Hz, and 8 000 Hz are investigated. The results show that the ability to integrate acoustic energy over time in patients with presbycusis is reduced compared to normals. It was possible to demonstrate quantitatively a relationship between the amount of hearing loss and the temporal integration. The difficulties in discrimination for patients with sensory-neural hearing loss may to some extent be explained from the reduced temporal integration.

Whenever a quantitative estimate of hearing loss is required, a standard pure tone audiometry based on tone pulses of 500 ms or more is performed. The spoken language contains many short and complex sounds thus the ability to hear these short sounds must also be of audiological interest.

The perception of short tones or temporal integration of acoustic energy can be measured by threshold determination of tone impulses of varying duration, i.e. short tone audiometry. Abnormal temporal integration has been considered as one of the reasons for reduced discrimination in hearing impaired ears (Hinchcliffe 1970; Broadbent & Stephens, 1970), but little information in this topic is available. This paper reports our results of short tone audiometry in 46 persons with presbycusis.

MATERIAL AND METHODS

This investigation is based on patients, age 53 to 86, with the diagnosis presbycusis and were selected among the patients in the ENT department and the Audiological Clinic, Gentofte Hospital.

The diagnosis was ensured by a typical history and otological and audiometric investigations. Subjects participating in this trial are present cases with mild as well as severe hearing losses.

Audiological equipment and testing procedure was the same as used in a previous study on normals (Pedersen & Elberling, 1972). The investigation is a monaural threshold determination using pure tone impulses of the frequencies 500 Hz, 1 000 Hz, 4 000 Hz and 8 000 Hz.

Equivalent energy time for stimulation was varied from about 2 ms to 1 000 ms. Prior to the investigation the subjects are informed about the nature of the test. Using the pure tone audiogram each threshold determination starts at a sensation level of 10 to 15 dB and the intensity is lowered in 2.5 dB steps.

At each frequency 10 different threshold measurements are performed corresponding to the 10 different impulse durations used.

Fig. 1 illustrates our method of calculating an estimate of the temporal integration (Pe-

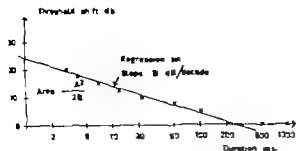


Fig. 1 Record from a test person. The measured thresholds at 1000 Hz. for 10 different stimulus durations. The figure illustrates our method to calculate the relevant expressions A , B and $-A^2/2B$ for the temporal integration.

densen & Elberling, 1972). In short the procedure is the following: the measured thresholds are plotted in a double logarithmic co-ordinate system, where the abscissa is the equivalent energy time of the stimulus, and the ordinate is the measured threshold shift in dB relative to the threshold for long stimulus (Benzon, 1953). Using the method of least squares a regression line is calculated through the measured points. This line has a slope of B dB/decade and cuts the ordinate axis in the A dB

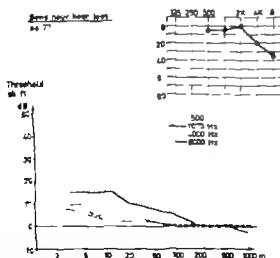


Fig. 2 The temporal integration in a case of mild presbycusis with high tone loss. The temporal integration (A , B) at 500 Hz and 1000 Hz are 22 and 27 i.e. normal. At 4000 Hz and 8000 Hz the temporal integration are 13 and 9 which is a significant reduction.

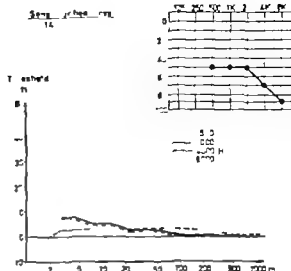


Fig. 3 The temporal integration of a severe case of presbycusis. The temporal integration at the frequencies 500 Hz, 1000 Hz, 4000 Hz and 8000 Hz are 18, 10, 10 and 6 respectively calculated by the formula $-A^2/2B$. This value is significantly lower than in normal persons.

point. The area of the triangle formed by the abscissa axis, the ordinate axis, and the regression line is calculated and used as a quantitative measure of the temporal integration. The area can be calculated with the knowledge of the A and B -value using the formula $-A^2/2B$.

RESULTS

Typical results from two subjects, a mild and a severe case of presbycusis, are shown in Figs. 2 and 3.

The results of the A -value, the B -value and the area $-A^2/2B$ were correlated to the actual hearing loss for each subject and each frequency. From Fig. 4 it is seen that the A value (threshold shift) is decreasing, when the hearing loss increases.

From Fig. 5 it is seen that the numeric value of B (slope of the regression line) is decreasing, when the hearing loss increases. The temporal integration expressed as $-A^2/2B$ decreases, when the hearing loss increases. This is illustrated for each frequency in Fig. 6.

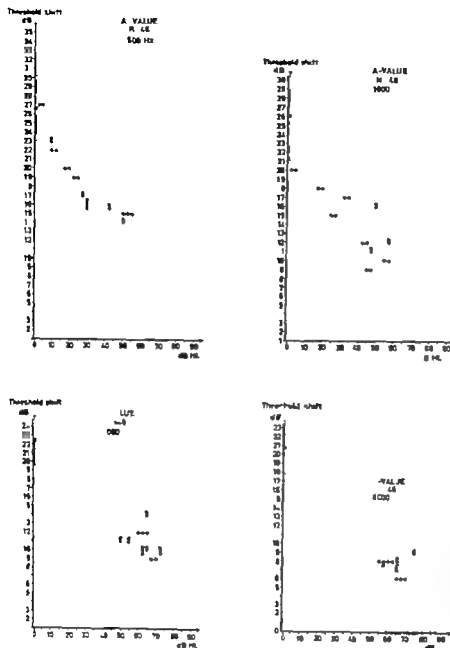


Fig. 4. The individual A-values (threshold-shift) versus the corresponding hearing losses at the 4 frequencies investigated. Mean values and standard deviations for normals are indicated.

DISCUSSION

In short tone audiometry on pathological ears Muskolczy-Fodor (1953) investigated 30 ears with perceptive deafness and recruitment. He demonstrated a decreased threshold shift for short tones in these patients compared with normals.

Harris et al. (1958) presented in a group of 25 patients, 10 cases of hearing defect due to

cochlear lesions. In most of these patients abnormal temporal integration was observed.

Elliot (1963) gives the first description of a connection between hearing loss and the degree of change in temporal integration based on a study of 36 patients with sensory-neural hearing loss.

Watson & Gengel (1969) and Gengel & Watson (1971) reported 11 cases of hearing impairment due to cochlear defects. In these pa-

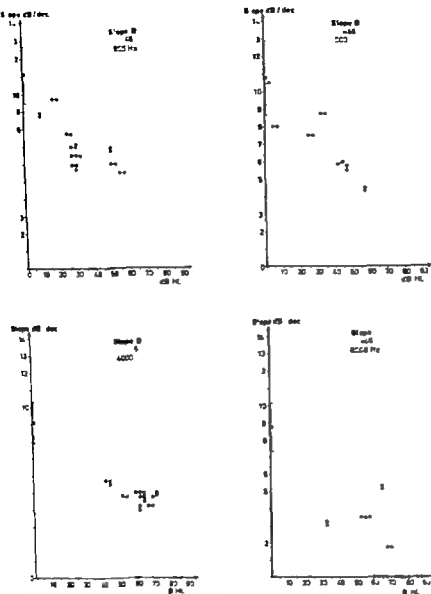


Fig. 5 The B-values (slope of regression line) versus the corresponding hearing losses at the 4 frequencies investigated. Mean values and standard deviations for normals are indicated.

tients they found a reduced ability of the auditory system to integrate over time the acoustic energy in brief sounds.

Sanders & Honig (1967) concluded on basis of results from 14 patients that ears with cochlear pathology is clearly distinguished from normal ears by short tone audiometry.

As a result of the investigations of Wright (1968) and Wright & Canelis (1969) it is concluded that in patients with mixed conductive sensory-neural component contributes to the change in temporal integration.

Most of the 46 persons with presbycusis investigated needed Hearing Aid (HA) and consulted the audiological clinic for this reason. To investigate patients with a minor degree of presbycusis a small number with a mild high-tone loss only was also included in the material. Care has been taken to exclude diseases other than presbycusis, but influence of acoustic trauma cannot be excluded completely.

From Fig. 4 it can be seen that the threshold shift (A-value) decreases, when the hearing-loss increases. Fig. 5 shows that the slope

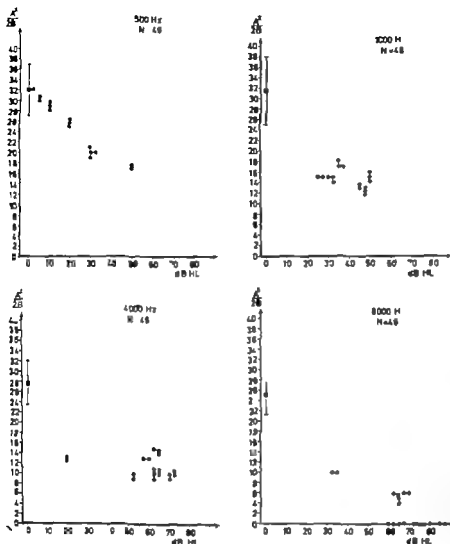


Fig 6 Temporal integration expressed as $-A^2/2B$, versus the corresponding hearing losses at the 4 frequencies investigated. Mean values and standard deviations for normals are indicated.

of the regression line is decreasing when the hearing loss increases. The temporal integration expressed by the value $-A^2/2B$ also shows a nearly inverse proportionality to the hearing loss as demonstrated in Fig. 6.

Of the expressions A , B and $-A^2/2B$ we think that $-A^2/2B$ is the most relevant for the temporal integration.

The dots indicating B -values show the great spread (Fig. 5) while the dots indicating values of A and $-A^2/2B$ appear with nearly identical spread (Figs. 4, 6). Of these two expressions $-A^2/2B$ is preferable, because changes in the B -value also is taken into account.

The results of short tone audiometry at 4

different frequencies in patients with presbycusis show that temporal integration of acoustic energy is reduced compared with normals (Pedersen & Elberling 1972), in agreement with the earlier investigations. It was possible to demonstrate quantitatively a relationship between the amount of hearing loss and the adequate expressions A , B and $-A^2/2B$ for temporal integration.

Difficulties in discrimination in patients with sensory-neural hearing loss have called for acoustological investigations (Hinchcliffe, 1970; Broadbent & Stephens, 1970). Our investigations confirm that in patients with presbycusis changes in thresholds produced by long at short acoustic stimuli are smaller than in pe

sons with normal hearing. If these patients are fitted with a HA, supposed to produce transient distortion (overshoot) changes in thresholds produced by long and short stimuli are perceived with an even smaller or negative difference. This might to some degree explain the difficulties in discrimination with sensory-neural hearing loss, even when supported with HA.

ZUSAMMENFASSUNG

Die Fähigkeit, akustische Energie über Zeit hin zu integrieren, d. h. die Temporalintegration, kann mit Kurztonaudiometrie untersucht werden. Veränderungen der Temporalintegration wurden als eine der Ursachen von Diskriminationserschwerigkeiten in schwerhörigen Ohren beobachtet. Die Ergebnisse einer Kurztonaudiometrie von 46 Presbycusis-Patienten werden hier mitgeteilt. Vier verschiedene Frequenzen (500 Hz, 1 000 Hz, 4 000 Hz und 8 000 Hz) sind untersucht worden. Die Resultate zeigen dass die Fähigkeit, akustische Energie zu integrieren, bei Presbycusis-Patienten reduziert ist, wenn man mit Normalgehör vergleicht.

Es war möglich, einen quantitativen Zusammenhang zwischen dem Grad des Hörverlustes und der Temporalintegration zu zeigen. Es ist wahrscheinlich, dass die mangelnde Temporalintegration bei Altersschwerhörigkeit in gewissem Masse die Diskriminationserschwerigkeiten dieser Patienten verursacht.

REFERENCES

Bentzen, O 1953 *Investigations on short tones*. Universitetsforlaget, Århus (Thesis).

- Brake Pedersen, C. & Elberling, C. 1972 Temporal integration of acoustic energy in normal hearing persons. *Acta Otolaryng* (Stockh) 74 398
- Broadbent, D. E. & Stephens, S.D.G. 1970 *Sensori neural hearing loss*. J. & A. Churchill Ltd., London (Ciba Symposium).
- Elliot, L. L. 1963 Tonal thresholds for short-duration stimuli as related to subject hearing level. *J Acoust Soc Amer* 35 578.
- Gengel, R. W. & Watson, C. S., 1971 Temporal integration. *J Speech Hearing Dis* 36 213
- Harris, J. D. Haines, H. L. & Myers, C. K. 1958. Brief-tone audiometry. *Arch Otolaryng* (Chic.) 67 699
- Hinchcliffe, R. 1970. *Sensori neural hearing loss*. J. & A. Churchill Ltd., London (Ciba Symposium).
- Miskolczy-Fodor F 1953 Monaural loudness-balance test and determination of recruitment degree with short sound impulses. *Acta Otolaryng* (Stockh.) 43 573
- Senders, J. W. & Honig, E. A. 1967 Brief tone audiometry. *Arch Otolaryng* (Chic.) 85 640.
- Watson, C. S. & Gengel, R. W. 1969 Signal duration and signal frequency in relation to auditory sensitivity. *J Acoust Soc Amer* 46 989
- Wright, H. N. 1968 The effect of sensori neural hearing loss on threshold-duration functions. *J Speech Hearing Res* 11 842.
- Wright, H. N. & Canella, F. 1969 Differential effect of conductive hearing loss on the threshold-duration function. *J Speech Hearing Res* 12 607

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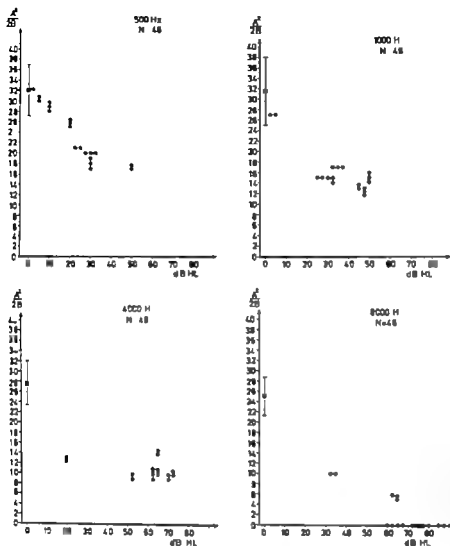


Fig. 6 Temporal integration expressed as $-A^2/2B$, versus the corresponding hearing losses at the 4 frequencies investigated. Mean values and standard deviations for normals are indicated.

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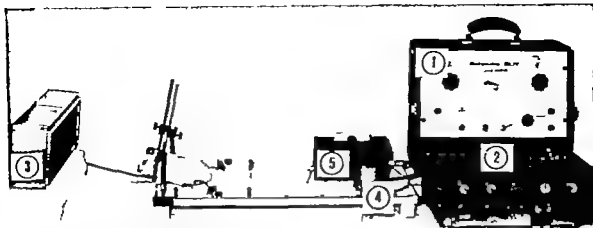


Fig. 1 Simplified equipment used for conditioning the guinea-pig to sound and for its Shiver-audiometry. 1. Audiometer; 2. amplifier; 3. loudspeaker (placed at exactly 20 cm from the animal's ear(s)); 4. sound

switch; 5. tension regulator with switch for sending the electric shock through the electrodes (e) attached to the hind-paws of the guinea-pig.

the experimental auditory damage caused by us with various methods.

MATERIALS AND METHODS

The original article by Anderson & Wedenberg should be consulted for details, the following is a report on the fundamental data of simplified S.A.

The first simplification concerns the use, as a source of pure tones, instead of the more complex electro-physiological equipment, of a common audiometer (mod. 802/A or mod. 200, Amplifon Milan), connected to an amplifier-loudspeaker system of appropriate power. This complex, which must be calibrated before each experiment, produces pure tones of 125 to 8000 Hz and of an intensity of -10 to +100 dB. It is used both to condition those animals which have already been subjected to a first selection and to carry Shiver-audiometry. Numerous preliminary experiments had allowed us to establish that the ideal experimental situation for us was to place the loudspeaker exactly 20 cm from the binaural axis of the guinea-pig, whose head—as in the original method—was held firm by a collar (Fig. 1).

A first examination of hearing based on the pinna reflex (acoustic reflex or Preyer reflex)

makes it possible to ascertain the intensity which should be adopted for the various frequencies from 250 to 8000 Hz tried out by us first for conditioning then for the audiometry.

Conditioning is obtained easily by means of the progressive presentation of pure tones at pre-determined intensities, each being followed at various intervals in order to avoid temporary conditioning, by a brief 2-5 V shock, applied through electrodes fixed to the animal's hind paws, by an appropriate tension regulator. During the conditioning phase, which takes place generally in 10-20 minute daily sessions, over a period of 3 to 8 days, a second selection of the animals takes place so far they had been selected merely on the basis of general conditions of health and of the normality of the tympanic membrane or of their evident incapacity for one reason or another to learn. In actual fact, the guinea-pig is easy to condition and it is clear that its aptitude for learning increases as the conditioning session progresses.

Another simplification introduced by us concerns the system for causing the shiver in the conditioned guinea-pig. Instead of the more or less complex systems of refrigeration—which are also, apart from anything else, a little noisy—we practise Shiver-audiometry in

a cold room intended for biochemical use thus the examination is conducted in the proper conditions of silence. Further advantages could derive from our present attempts to use a system similar to that of common cryostats. When placed in the cold room (0–2°C), after 10 to 20 minutes the guinea-pig shows a clear shiver. This, as well as its interruption by an appropriate acoustic stimulus and in conformity with Anderson & Wedenberg, can easily be recorded together with the sound stimulus by using a piezoelectric microphone connected to an ordinary two-channel cardiograph. However we prefer not to record the shiver just as we have abstained from any possible automation of the conditioning phase because we believe that it is very important that each phase of the technique be directly followed by the examiner.

The values in dB of the minimum intensity which for the individual frequencies between 250 and 8000 Hz, cause the interruption of the shiver correspond to the hearing threshold of the animal, and are marked on an ordinary human audiometric chart. In experiments on auditory pathology the initial normal values are set to zero and thus any pathological values can be appropriately corrected. On the chart we also always register the threshold values obtained for the same scale of frequencies with pinna reflex because as Anderson & Wedenberg have shown they are an indication of lesions to the receptor in other words of recruitment, if they remain more or less unchanged when the S.A. reveals an auditory deficit.

Shiver-audiometry has been conducted both on albino guinea-pigs (Morini farm, R. Emilia, Italy) and on pigmented guinea-pigs of both sexes and of an age varying from 2 weeks to about 2 years. Normal auditory values have been obtained from guinea-pigs of about 45–60 days, corresponding to a body-weight of about 200–350 g. During a second phase certain guinea-pigs which, according to S.A. had normal hearing were used for pathological experiments.

Four albino guinea-pigs were treated subcutaneously four times a day with a dose of 250 mg per kg of body weight per day of sulphate kanamycin for 20 days. The behaviour of their hearing was studied every 10 days, at first in all 4 animals then after the interruption of the treatment, only in 3 of them one having died on the 19th day.

In 4 albino and 4 pigmented guinea-pigs we studied auditory behaviour after an hour of hyperstimulation with a noise of a frequency band of 80 to 8000 Hz, and of an intensity at the level of the guinea-pig's auricle and in our experimental conditions, equal to 126 dB (phonometer mod. 908, Amplifon, Milan). Shiver-audiometry was carried out after 1, 2, 5 and 8 days.

Finally in order to evaluate whether S.A. could be used in experiments based on endo-tympanic injection treatment, 4 guinea-pigs under ether narcosis were injected transtympanically with about 0.20 ml of a saturated sodium chloride solution in order to obtain a Ménière-like syndrome as described by Arslan (1969). Four other albino guinea-pigs of the same age acted as controls and were treated in the same manner with physiological solution. These were indispensable for as is well known, any liquid which is introduced into the guinea-pig's middle ear causes, on the one hand, the rapid disappearance of the pinna reflex and on the other otitis. In an attempt to prevent otitis, the animals were treated with an intramuscular injection of 10 000 U.O. of sodium penicillin in 0.1 ml of distilled water at intervals of 12 hours. S.A. was conducted on the 7th day.

The data obtained from audiometric experiments have been elaborated statistically with an evaluation of the standard error of the average and an analysis of the variance.

RESULTS

Table I gives the average values and the standard deviation of the average obtained for the frequencies of 250 to 8000 Hz in the audiomet-

Table I Average auditory threshold (\pm E.S.) expressed in dB for the frequencies of 250–8 000 Hz, obtained by Shiver-audiometry in albino and pigmented guinea-pigs

Guinea pig	No. of animals	250	500	1000	1500	2000	3000	4000	6000	8000 Hz
A. Albino	31	0.1 ± 1.6	2.3 ± 1.1	4.7 ± 1.1	3.7 ± 1.5	2.7 ± 1.2	1.5 ± 1.3	-1.7 ± 1.4	-7.1 ± 1.3	-3.9 ± 0.7
B. Pigmented	5	25.0 ± 1.5	28.0 ± 1.2	28.0 ± 3.0	28.4 ± 2.6	33.0 ± 2.0	29.4 ± 1.7	28.0 ± 1.5	20.8 ± 1.6	28.0 ± 2.0

A B $P < 0.001$ for all frequencies.

ric examination of 31 albino and 5 pigmented normal guinea-pigs in our experimental conditions.

In all the frequencies examined, a statistical comparison of the two groups has revealed a significant difference ($P < 0.001$) from which it may be deduced that in our experimental conditions the albino guinea-pig has a better auditory capacity than the pigmented one. The reason for this, for the time being, is a mystery to us.

Fig. 1 shows the average threshold curves obtained by S.A. from 20 albino guinea-pigs which, after all the usual checking operations, were considered to be normally-hearing, whereas in actual fact 4 of them had hypoacusia in almost all the frequencies examined. It

may be observed that this deficit was not revealed by the examination carried out with the pinna reflex. An anatomical examination of these animals showed that they were suffering mildly from unilateral or bilateral otitis. Shiver-audiometry therefore makes it possible to identify those guinea-pigs which really do have normal hearing, while any judgment as to normality which is based on general conditions of health, on the condition of the tympanic membranes and the presence of the pinna reflex could lead to erroneous selection.

Fig. 3a and b show the behaviour of hearing during subcutaneous treatment with high doses of kanamycin. Since one of the guinea pigs died towards the end of the treatment, Fig. 3b gives the average auditory curve of 3 animals only as obtained after the end of the treatment.

During the treatment we were able to detect a progressive auditory deficit, especially at high and low frequencies, while the 1 500 Hz frequency appeared to remain even. After suspension a recovery of the lower frequencies was detected, whereas the deficit of the higher frequencies persisted, and even the middle frequencies were involved.

The final result was a descending curve towards the high tones. The persistence of the pinna reflex during and after the kanamycin treatment indicates that the damage affects the receptors. Observations during the first phase of the experiment are curiously in harmony with the data of other authors who, by means of the cochleogram technique, have studied the neurosensorial lesion which can thus be pro-

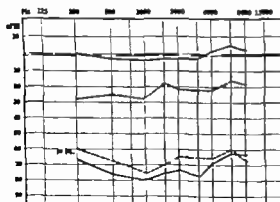


Fig. 2 Average curves of auditory threshold obtained with Shiver-audiometry in 16 albino normally-hearing guinea-pigs (—) and 4 albino guinea-pigs of the same group which, though apparently normal in preceding tests proved to have hypoacusia (- -) P.R. average threshold curve obtained with the pinna reflex.

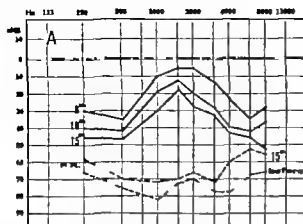
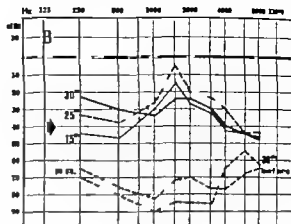


Fig 3 (A) Average hearing threshold curves in relation to the initial average values set to zero and obtained by Shiver-audiometry in 4 albino guinea-pigs during subcutaneous treatment with kanamycin (250 mg/kg of body weight/day). Examination shows a progressive auditory deficit for the frequencies of 500–1 000 Hz and 3 000–8 000 Hz. The behaviours of



the pinna reflex (P.R.) indicates that lesion is in the receptors.

Fig 3 (B) After twentieth day (arrow) and suspension of the treatment, progressive recuperation may be observed in the 3 surviving animals for the lower frequencies, while the middle ones are involved and the higher ones continue to show a deficit.

voked (Ostyn & Tyberghien, 1968), whereas the deficit observed after the treatment is reminiscent of that to be met with in patients suffering from antibiotic intoxication.

Fig. 4a and b refers to the audiometric results obtained with albino and pigmented guinea-pigs after acoustic hyperstimulation (white noise of 126 dB for an hour). From the average threshold curve obtained at various intervals

from the trauma, it can be seen how immediately after it, the deficit is extremely important and almost equal for all the frequencies in the case of the albino guinea-pig, whereas it is more important in the medium frequencies in that of the pigmented animal. Moreover the behaviour of the pinna reflex during the experiment shows that here we have damaged receptors. But in the following phases of the

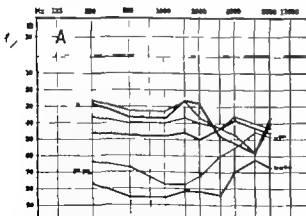
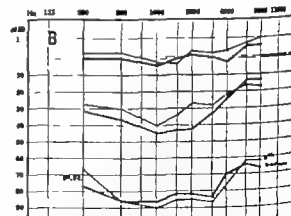


Fig 4 (A) Evolution of the average auditory threshold curves (4 animals) obtained after 4 hours, 2, 6, 8 days from an acoustic overstimulation (white noise 126 dB 1 hour) upon albino and pigmented guinea-pigs. (A) Albino guinea-pig. Compared with the normal average values set to zero, after a considerable initial deficit a progressive recovery may be ob-



served for the 250–2 000 Hz frequencies and a further drop for the 3 000–6 000 Hz frequencies. The behaviour of the pinna reflex (P.R.) indicates a lesion of the receptors. (B) Pigmented guinea-pig. As early as the sixth day there is a complete recovery of the normal auditory functions.

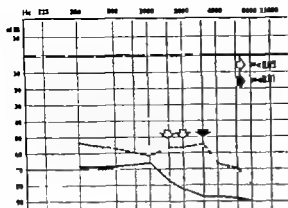


Fig. 5 Average curves of the hearing threshold, in relation to the normal values set to zero, obtained with Shilver-audiometry on the seventh day after the endotympanic injection of a saturated sodium chloride solution (—) and, respectively of a physiological solution (---). The animals of the first group reveal a deficit for the 1 500–3 000 Hz frequencies.

experiment, the behaviour of hearing in the two experimental groups is entirely different. On the 8th day the albino guinea-pig showed a low degree of recuperation, limited to the low and medium frequencies while the deficit in the higher frequencies was more pronounced. The pigmented guinea-pig on the contrary revealed even on the 6th day a complete recuperation for all the frequencies examined.

This observation appears to be in agreement with other researchers (Galloto & Bonaccorsi, 1965) who had registered by a different method a weaker resistance of the albino guinea-pig's ear to the acoustic trauma, and thus appears to be a new proof in support of Cherubino's hypothesis (1968) that the pigment itself in conditions of hypoxia caused by the acoustic trauma, could well be a source of metabolic energy for functional recuperation. Such recuperation has been shown by present experiments to be only partial twenty days after the acoustic trauma as far as the albino guinea-pig is concerned.

Finally Fig. 5 shows the behaviour of the hearing of the albino guinea-pig 7 days after a bilateral endotympanic injection of a saturated sodium chloride solution. Comparisons

were made with guinea-pigs treated with physiological solution. As statistic analysis has shown, there is an important deficit only for the frequencies between 1 500 and 3 000 Hz ($P < 0.05$ or 0.01). Since the endotympanic injection leads to the disappearance of the pinna reflex, we are not able to ascertain audiotically whether it is a question of a deficit of the receptors, but the morphologic controls in our possession (unpublished data) show that as a result of endotympanic treatment with saturated sodium chloride solution there arises a serious alteration of the permeability of the membranous labyrinth, this being accompanied for the first hour by a general swelling of the dendrites of the internal hair cells. This lesion which is itself specific in that it has been met with in various experimental conditions (Spöndlin, 1970) proves that there is also sensori-neural damage. Moreover our audiometric results seem to agree with those obtained from cats under the same experimental conditions by means of other tests by Molinari (1971), who noted a deficit in the middle frequencies.

CONCLUSIONS

Since most of experimental otologic research is conducted on guinea-pigs, a technique which allows the majority of research workers to evaluate the behaviour of hearing in this animal is normal and in provoked pathological conditions can be important.

The simplification of the technique proposed by Anderson & Wedenberg as applied by us, furnishes a method which, in accordance with these authors' reports, fully corresponds to its purpose. Simplified S.A. is actually easy to practise insofar as it does not require complicated equipment but only common and easily obtainable instruments. Above all, compared with all other methods proposed so far it is the least expensive of tests and that which can be put to the most varied uses. The experiments given by us as examples show clearly how versatile it can be. It makes possible the identification of truly normally-hearing guinea

pigs, and indicates that in our experimental conditions the best animal for research work is the albino guinea-pig. Moreover it has made it possible to follow up in time the evolution of hearing in experiments of short and medium duration and given us indications, through the deficits observed as to the cochlear sectors which deserve morphological study. Also as experiments now being conducted have shown, the reconditioning of the animal might permit the prolongation of observation which at present lasts little more than 2 months, whereas it is possible to use this method on very young guinea-pigs whose audiogram could be obtained as early as their tenth day of life. The test based on the pinna reflex—which some investigators still use—has shown its inadequacy in evaluating the behaviour of hearing in the guinea-pig whereas Shiver-audiometry has been extremely useful in following up dynamically any possible auditory damage and in evaluating the difference in behaviour between the albino and the pigmented guinea-pig after acoustic hyperstimulation. Another advantage could be—when using the proper controls and statistic analysis—the possibility to study the effects caused by endotympanic treatment. We consider that these possible applications are well worth further study and are carrying this further at present. Certain pressing problems of otology can be dealt with more completely such as for instance the problem of identifying ototoxic drugs, this method having proved capable of establishing the extent and the seat of the damage obviously this can be useful for precise evaluation in relationship to dosing, means of application, rhythm and duration of the ototoxic treatment. For all these reasons, Shiver-audiometry may be recommended as the simplest, most practical and least expensive of tests for the study of hearing in the guinea-pig this animal being the most extensively used in otological experimental research.

ACKNOWLEDGMENT

The author wishes to thank Miles P. Settembrini and Dr A. Sidoli for their technical cooperation

ZUSAMMENFASSUNG

Der Hörversuch von Anderson und Wedenberg ist hier vereinfacht und wegen des Bestands, der seine Originalität bildet, Schanderaudiometrie genannt worden. Da seine Benützung nun einfach und billig geworden ist, zeigt sich auch seine Vielseitigkeit, was es die Versuche mit albinistischen und pigmentierten Meerschweinchen beweisen. Dank dieser Versuche konnte man Tiere mit Normalgehör identifizieren und dadurch feststellen, dass in unserem Versuchszusammenhang der Albino eine größere Hörfähigkeit als das pigmentierte Tier besitzt. Das Albino-Meerschweinchen weist auch eine größere Verwundbarkeit der Ohrschnecke unter akustischer Hyperreizung auf. Die Schanderaudiometrie hat sich bei Versuchen von kurzer und mittlerer Dauer als ein wichtiger dynamischer Indikator der Gehörentwicklung erwiesen, da sie nicht nur die Entwicklung der Läsionen in der Zeit, sondern auch ihr Ausmass feststellt. Sie ist also sehr nützlich, da sie der morphologischen Forschung den Weg zu den am meisten verletzten Sektoren der Ohrschnecke zeigt, z.B. in Fällen von Kanamycyloempfindung oder nach akustischem Trauma oder nach lokaler Behandlung des Ohrs mit einer gestützten Othrinatriumlösung. Auf Grund dieser Eigenschaften bildet die Schanderaudiometrie eine wertvolle Vervollständigung der experimentellen otologischen Forschung.

REFERENCES

- Anderson, H. & Wedenberg, E. 1965 A new method for hearing test in the guinea-pig. *Acta Otolaryng (Stockh.)* 60 375
- Arlan, M. 1969 Modifications of the osmotic pressure of perilymph and endolymph. *Acta Otolaryng (Stockh.)* 67 360
- Cherubino, M. 1968. I melanociti dell'orecchio interno. *Atti Accad. Med. Lomb.* 23 20.
- Crifò, S., Settembrini, P. & Sidoli, A. 1970. Britido-anestesia della cavia. *Il Valmista* 46 286.
- Galloto G. B. & Bonaccorsi, P. 1965 Il fattore costituzionale del trauma acustico individualizzato apertamente nella diversa concentrazione di melanina nella stria vascolare. *Boll. Med. Orecch.* 78 1289
- Molinari, G. 1971 *Effects of osmotic pressure in the vestibular receptors* Paper read at 8th Workshop on Inner Ear Biology Venice.
- Ostyn, F. & Tyberghele, J. 1968. Influence of some streptomycetes antibiotics on the inner ear of the guinea-pig. *Acta Otolaryng (Stockh.)* Suppl. 214
- Spoendlin, H. 1970. Auditory vestibular olfactory and gustatory organs. In *Ultrastructure of the peripheral nervous system and sense organs* (ed. J. Bialhoff) G. Thieme, Stuttgart.

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DER SUCCINATDEHYDROGENASE-NACHWEIS MIT HILFE DER PERFUSIONSTECHNIK AN DER COCHLEA DES MEERSCHWEIN- CHENS UNTER NORMALBEDINGUNGEN UND NACH DAUERLÄRMEINWIRKUNG

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(Eingegangen am 6. März, 1972)

Abstract. Mit Hilfe der Perfusionstechnik wurde die SDH in der normalen Meerschweinchencochlea nachgewiesen. Abgesehen von dem Fehlen der Färbung in der Stria vascularis und in den Ganglienzellen des Ganglion spirale entspricht die Verteilung der Aktivität im wesentlichen den Ergebnissen bei der vielfach verwendeten Schnitt- und Stückinkubation. Nach 63 Stunden Einwirkung von 110 dB „weißem Rauschen“ (9 h Lärm, 15 h Pause, Samstag und Sonntag Pause) kommt es zu einem eindeutigen Abfall der SDH in den äußeren und inneren Haarzellen und den zugehörigen Nervenendigungen. Eine unschriebene Lokalisation des Aktivitätsabfalls auf ein bestimmtes Areal des Ductus cochlearis konnte nicht gesichert werden. Die Ergebnisse werden diskutiert.

Entsprechend der hervorragenden Bedeutung der Succinatdehydrogenase (SDH) im Energiestoffwechsel der Zelle wurden im Rahmen der histochemischen Untersuchung des Innenohres häufig Arbeiten über ihre Verteilung in der Cochlea publiziert. Zahlreichen licht und elektronenmikroskopischen Studien nach Stück und Schnittinkubation (Vosteen 1956 a b c 1960 1961 Kolde et al., 1962 Nakai, 1965 Nakai & Hilding, 1968) stehen relativ wenige Beobachtungen nach Perfusionsinkubation gegenüber (Gehrhardt, 1961 a b Schätzle & Müsebeck, 1962, Spendlin & Balogh, 1963 n b 1964).

Unterstützt durch einen Forschungsauftrag des Ministeriums für Gesundheitswesen der DDR.

Da nach unserer Meinung der Perfusionstechnik bei Untersuchungen des Cortischen Organs eine größere Bedeutung zukommt, als ihr momentan zuerkannt wird, fühlen wir uns berechtigt, unsere Ergebnisse, die im Rahmen methodischer Vorarbeiten zu Studien und Veränderungen nach Lärmbelastung durchgeführt wurden, zu publizieren.

MATERIAL AND METHODE

Insgesamt wurden 18 Meerschweinchen (250-500 g Gewicht) beiderlei Geschlechts mit normalem Preyer Reflex untersucht. In Urethan-Ather-Narkose erfolgte die Präparation und Eröffnung der Bulla tympanica sowie die Freilegung des runden und ovalen Fensters. Nach Anlegen eines apicalen Bohrloches in der Schnecke wurde ein PVC Schlauch in das runde Fenster eingekittet und das Inkubationsgemisch perfundiert.

Der SDH-Nachweis erfolgte mit dem Medium von Ogawa & Barnett (1965) mit TNBT als Indikator bei einer Temperatur von 60 Mm. Sofort nach der Inkubation wurde mit neutralem, exgekühltem, 4%igem Formalin mit einem Zusatz von 7,5% Sacrose (Geyer 1969) 10 Min lang per perfusionem vorfixiert.

Nach Dekapitation und Isolierung der Cochlea aus dem Felsenbein erfolgte im gleichen Medium die Nachfixierung 16 h lang bei 4 C.

Die Entkalkung wurde in neutraler gesättigter Dinatrium EDTA-Lösung (mod. n. Balogh, 1965) die durch eingeleitete Luftblasen ständig bewegt wurde, 7 Tage lang bei 20 C durchgeführt. Danach wurde in Polyäthylenglycol 600 nach Halbhuber & Geyer (1966) eingebettet, am Gefriermikrotom etwa 15 µm dick geschnitten und in Glyceringelatine eingedeckt. Als Kontrollversuche inkubierten wir nach Geyer (1969)

- unter Weglassen des Substrats
- nach Praeinkubation mit 4% Formalin, 30 Min bei 20 C
- substratfrei nach 30 Min. Auswaschen des endogenen Substrats mit 0.44 M Sucrose-lösung
- unter Zusatz von 1,5 ml 0.1 M Natrium malonat zum Inkubationsmedium.

Die Beschallung von insgesamt 6 Tieren erfolgte nach Dieroff (1971) sowie Dieroff & Beck (1966) durch Einwirkung von „weißem Rauschen“ mit einer Intensität von etwa 110 dB 63 h lang (9 h tägl., 5 Tage hintereinander Samstag und Sonntag Pause, danach nochmals 2 Tage lang) 15 h nach der letzten Beschallung wurde die histochemische Untersuchung begonnen.

ERGEBNISSE

Normales Cortisches Organ

Im normalen Cortischen Organ wurden die stärksten SDH-Aktivitäten in den inneren und äußeren Haarzellen nachgewiesen (Abb 1a) wobei die Formazanablagerungen in den inneren Haarzellen stets intensiver schienen.

Die Farbniederschläge in den äußeren Haarzellen waren in der Intensität abgestuft (Abb 1b) wobei ein kräftiges Depot in der Intermediärzone (Schema nach Iurato 1961) von einem infranukleären, noch intensiveren For-

mazanlager zu unterscheiden war. Im Bereich der perinukleären Zone fand sich eine deutliche farbliche Betonung der zellwandnahen Bezirke und gegenüber den erwähnten Depots in der Intensität abfallende im übrigen Cytoplasma unter Aussparung des Kerns verteilte Farbablagerungen. Die inneren Haarzellen (Abb 1c) zeigten eine derartig deutliche Abstufung in der Färbung nicht. Die Farbniederschläge waren granulär sie lagen relativ gleichmäßig verteilt unter Aussparung des Kerns im gesamten Cytoplasma. Nur bei gelegentlich etwas schwächerer Gesamtfärbung fanden sich deutlichere Depots in der Intermediären Zone und dem infranukleären Bereich.

In der Zone der Nervenendigungen am unteren Zellpol der äußeren Haarzellen (Abb 1a 2) wurden kräftige Ablagerungen von Farbstoff gefunden, die von den kappenförmigen infranukleären Depots deutlich abgrenzbar und auch weniger intensiv waren. Von diesem Bereich zogen sich entlang der Randbezirke der Deiters-Zellen in Richtung Basalmembran kleinere, perlschnurartig aneinander gereihte Farbstoffpunkte (Abb 2). Es konnte auch bei maximaler Vergrößerung nicht sicher zwischen extrazellulärer und intrazellulärer Lage entschieden werden. Bei Darstellung des Tunnelspaltbündels (Abb. 1a 1c) im Schnitt wurde es bei genügender Vergrößerung immer deutlich angefärbt gefunden. Gelegentlich war auch die Tunnelfaser positiv.

Die Zone der Nervenendigungen unter den inneren Haarzellen (Abb 1a, 1c) zeigte ebenfalls intensive SDH-Aktivitäten, wobei die Formazandepots deutlich kelchartig den unteren Zellpol bis in Höhe des Zellkerns hinauf umgriffen. Von dort zogen sich die Farbstoffablagerungen (Abb 1a, 1c) kontinuierlich in gleicher Stärke bis zur Habenula perforata hin. Modioluswärts setzten sie sich dann unter regelmäßigem und stetigem Intensitätsabfall entlang der Nervenfasern fort. Weiter zur Nähe des Ganglion spirale hin konnten noch immer vereinzelt Ansammlungen von Formazangranula im Verlauf der Nervenfasern nach-



Abb 1 SDH-Reaktion am normalen Corti-Organ des Meerschweinchens. (a) Massive Reaktion in äußeren und inneren Haarzellen, jeweils eingeschlossen die Zone der Nervenzellnerven. Besonders zu beachten das positive Transversalspiralbündel (Tsp). 600 (b) Äußere Haarzellen mit typischer Verteilung der Formazandepots. 1 500. (c) Innere Haarzellen, charakteristische Darstellung der Zone der Nervenzellnerven. Anfärbung der Nervenfasern positives Transversalspiralbündel. $\times 1 500$.

gewiesen werden. Selbst im intraganglionären Spiralbündel wurden sie gelegentlich gefunden.

Die Nervenzellen des Ganglion spirale bleiben eindeutig negativ

Im Zellkörper der Deltazellen, abgesehen von den beschriebenen randständigen perlschnurartigen Farbstoffdepots, wie auch in den übrigen Stützzellen des Cortischen Organs



Abb 2 SDH Reaktion im Striazellbereich unter den äußeren Haarzellen. Positive Nervenzufügungen, Anfärbung der äußeren Spiralfasern, Deiterszellen separat 1500.

und in den Boettcher Zellen konnten keine Aktivitäten nachgewiesen werden.

Mehr oder weniger vereinzelt und unregelmäßig verteilt wurden SDH Aktivitäten in den Interdentalzellen gefunden (Abb 3 a 3 b) die



Abb 3 SDH Reaktion an den Interdentalzellen der normalen Meerschweinchencochlea. (a) 4 positive Interdentalzellen zwischen mehreren negativen. 610 (b) Deutliche Reaktion im gesamten Phalanxbereich bei fehlender Färbung im vaskulären Hohlraumreich. 1440.

wenn positiv recht kräftige Farbstoffablagerungen zeigten. Dabei ließen sich immer im gesamten cytoplasmahaltigen Zellleib unter Aussparung des Kerns bei farblicher Betonung des supranukleären Zellteils SDH Aktivitäten nachweisen. In den Fällen in denen der „intracytoplasmic duct“ (Lim, 1970 b) in gleicher Ebene mit dem übrigen Zellteil dargestellt werden konnte (Abb. 3 b) zeigte sich eindeutig das Fehlen von Formazangranula in dieser intrazellulären Hohlraumbildung. Eine geringe, aber stetige SDH Aktivität zeigte sich im bindegewebigen Anteil des Lumbus spiralis.

Die Stria vascularis blieb in allen Präparaten durchweg negativ. Das Ligamentum spirale zeigte besonders hinter dem Epithel der Prominentia spiralis und oberhalb des Ansatzes der Reißner Membran eine eindeutige Färbung, sowie eine deutliche, wenn auch weniger intensive Färbung in allen übrigen Bereichen wobei vereinzelte, fibrocytenähnliche Zellen durch stärkere Farbstoffablagerungen hervorgehoben waren.

Im Epithel des Sulcus externus und der Prominentia spiralis konnten keine Aktivitäten nachgewiesen werden.

In den Kontrollversuchen mit Formolpräinkubation zeigte sich lediglich im Bereich des



Abb 4 SDH Reaktion am Corti-Organ des Meerschweinchen nach Einwirkung von weißem Rauschen 63 h mit 110 dB. (a) Abschwächung der Reaktion in inneren u. äußeren Haarzellen. 570 (b) Äußere Haarzellen mit Erhaltung der Depots in der Zona intermedia u. den infranucleären Zellbereichen, Nervenendigungen positiv 1420. (c) Innere Haarzelle. Erhaltung des Depots in der Zona intermedia, Nervenendigungen und Nervenfasern positiv 1420, Spiralfaserbündel ebenfalls positiv 1420.

amentum spirale mit Ausnahme der Ge-
gend hinter der Prominentia spiralis und ober-
halb des Ansatzes der Reißner-Membran eine
feuchtere Farbstoffablagerung. Auch nach
Substratauswaschung mit 0,44 M Sacroselö-

sung und substratfreier Inkubation blieb noch
ein positiver Anteil in dem beschriebenen Be-
reich zurück. Nach Malonat-Hemmung wurde
jedoch auch dieser Teil völlig negativ. Alle
übrigen ausgewerteten Strukturen blieben bei

den durchgeführten Kontrollversuchen ohne Reaktion.

Dauerlärmeinwirkung

Die SDH Reaktionen nach Dauerlärmeinwirkung zeigten in den verschiedenen Präparaten relativ gleichförmige Ergebnisse. In den inneren und äußeren Haarzellen, sowie den dazugehörigen Nervenendigungen und Nervenfasern konnte eine eindeutige Abschwächung der Färbung festgestellt werden (Abb. 4 a b c). Hinsichtlich der intrazellulären Verteilung kam es zu keinen eindeutigen Veränderungen. So blieben bei einer Abnahme der Reaktion in den äußeren Haarzellen insgesamt sowohl die Zona intermedia als auch die infranukleären Zellbereiche und die randnahen Bezirke der perinukleären Zone in den entsprechenden Relationen betont (Abb. 4 b). Die zugehörigen Nervenendigungen und die beschriebenen perlschnurartigen Farbstoffdepots am Rande der Deiters-Zellen waren deutlich schwächer gefärbt als im normalen Corti-Organ (Abb. 4 a). Die inneren Haarzellen waren hinsichtlich ihrer SDH Aktivität etwa im gleichen Maße abgeschwächt (Abb. 4 c). Auch hier blieb eine farbliche Betonung der Zona intermedia bestehen. Die Nervenendigungen unter den inneren Haarzellen färbten sich ebenfalls weniger intensiv. Gleiches gilt für die beschriebenen Aktivitäten im Bereich der Nervenfasern in der Lamina spiralis ossea.

Die übrigen bei der Auswertung der normalen Cochlea als negativ oder positiv beschriebenen Strukturen erfuhren hinsichtlich ihrer Färbungsintensität keinerlei Veränderungen. Ein Unterschied der Abnahme der SDH Aktivität im Cortischen Organ nach der Lärmeinwirkung der verschiedenen Windungen zueinander konnte nicht eindeutig nachgewiesen werden.

DISKUSSION

Die Anwendung der Perfusionsmethodik zum histochemischen Nachweis mitochondrial gebundener Enzyme bei der Untersuchung von

Veränderungen nach Lärmeinwirkung auf der Ebene der Stoffwechselvorgänge in der Cochlea bietet u. E. eine Reihe von Vorteilen gegenüber anderen Methoden.

Die Diffusion des Inkubationsmediums und seine Reaktion mit dem Enzym erfolgt unter intravitalem Bedingungen. Daher ist die Anschaltung ortsunrichtiger Ablagerungen von Farbstoffdepots durch Enzymdiffusion bei gebundenen Enzymen weitgehend möglich. Außerdem kann nach Beendigung der Reaktion die Fixation des Gewebes praktisch *intra vitam* erfolgen. Dadurch und im Verein mit der Anwendung neuerer Kunststoffeinbettungen anstelle des ungünstigen Gelatineverfahrens kommt es zu verbesserter Strukturhaltung, die günstigere Bedingungen für die Auswertung und Dokumentation der Versuchsergebnisse schafft.

Unsere Befunde stimmen im wesentlichen mit den Untersuchungen Spoendlin & Balogh (1963 a b 1964) überein. Auch in unseren Schnitten blieben die Stria vascularis und das Ganglion spirale eindeutig negativ obwohl sie SDH in großen Mengen enthalten (u. a. Votaw 1956 a b c). Spoendlin & Balogh erklärten diesen Befund mit einer Diffusionsbarriere zwischen Ligamentum spirale und Stria vascularis, die die freie Diffusion des Inkubationsmediums aus den weiten interzellulären Spalten des Ligamentum spirale in das Gewebe der Stria vascularis hinein behindert. Durch die eindrucksvollen Untersuchungen der Transportvorgänge in der Cochlea mit Thorotrast durch Ilberg (1968 a) konnte diese Vermutung bestätigt werden. Danach waren die Interzellularräume der Stria vascularis sowohl gegen die Endolymph der Scala media als auch gegen das Ligamentum spirale hin durch Zonula occludentes verschlossen, wodurch die freie Diffusion der als Tracer verwendeten Moleküle verhindert wurde.

Die von Gehrhardt (1961 a, b) untersuchten Cochleae zeigten demgegenüber eine intensive Farbstoffablagerung in der Stria vascularis und den Zellen des Ganglion spirale. Diese Diskrepanz zu unseren Befunden und den Er-

gebissenen Spoendlin & Balogh (1963 a b 1964) läßt sich offenbar durch das im Laufe der Vorbereitung zur Inkubation durchgeführte Einfrieren und Wiederauftauen erklären. Dabei kommt es nach Geyer (1969) zu Veränderungen an den Membranen die die Diffusionsbedingungen erschweren. Auf diese Weise könnte eine Überwindung der durch Ilberg nachgewiesenen Penetrationsbarriere möglich werden. Die Gefahr der Enzymdiskontamination ist dabei jedoch erheblich.

Für die Positivität der *Stria vascularis* in den ebenfalls mit Hilfe der Perfusionstechnik durchgeführten Untersuchungen von Schätzle & Müsebeck (1962) können wir zunächst keine Erklärung finden.

Das Fehlen von Farbstoffablagerungen in den Zellen des Ganglion spirale unserer Präparate muß zunächst ebenfalls mit Behinderung der ganglionwärts gerichteten Ausbreitung des Inkubationsmediums erklärt werden.

Der Nachweis von SDH Aktivitäten im Nervus acusticus mit modioluswärts abnehmender Intensität und geringer aber eindeutiger Formazandepots im weiteren Verlauf des Nerven bis hin zum Ganglion spirale unter Einbeziehung des Intraganglionären Spiralbündels weist jedoch auf eine begrenzt durchlässige Verbindung zwischen Cortilymphraum und Ganglion spirale hin.

Arnold & Ilberg (1971) fanden einen solchen Diffusionsweg nach Injektion von Thorotrast in die Cisterna cerebellomedullaris entlang des Perineuralspaltes des Nervus acusticus, der nach Untersuchungen von Shanta & Bourne (1963) mit einem mehrschichtigen, endothelähnlichen Epithelschlauch gegen das perineurale Bindegewebe abgegrenzt wird.

Die intensiven Farbstoffablagerungen in ver einzeln Interdentalzellen weisen auf eine erhebliche SDH Aktivität hin. Beim Nachweis von NADH Diaphorase fand Lim (1970 a) ebenfalls massiv positive neben völlig negativen Zellen was der Autor mit unterschiedlichen Stoffwechselniveaus erklärte, die durch mitlerende Faktoren der Glykolyse reguliert werden könnten. Nach ultrastrukturellen Be-

funden (Lim, 1970 b) sind in Interdentalzellen mit intracytoplasmatischem Gang u. a. die Mitochondrien zahlreicher und besser entwickelt als in solchen ohne Gang.

Die positiven Interdentalzellen unserer Präparate zeigen teilweise unabhängig vom Zellkern größere Aussparungen des ansonsten in allen Anteilen einschließlic Phalanx (Lim, 1970 b) stark gefärbten Zellkörpers, die den beschriebenen Gängen entsprechen könnten. Da über die zur Membrana tectoria hin gelegene Öffnung des „intracytoplasmic duct“ Sekretion von Material beobachtet wurde (Lim, 1970 b) das nach Ansicht mehrerer Autoren für die Membrana tectoria bestimmt ist (Belanger 1953 Iurato, 1962 Ilberg, 1968 b Lim, 1970 b) könnte es sich hier um einen durch verschiedenen starke Sekretionsstärke bedingten unterschiedlich hohen Verbrauch an Energie und Grundbausteinen handeln.

Das Inkubationsmedium mußte entsprechend der von Ilberg (1968 b) verfolgten Thorotrastdiffusion aus der Perilymphe der Scala tympani und vestibuli in die Interzellularräume des Limbus spiralis und von dort ohne Behinderung durch die Basalmembranen (Ilberg, 1968 b) in die Interdentalzellen gelangen.

Die geringen, aber regelmäßigen SDH-Aktivitäten im bindegewebigen Anteil des Limbus spiralis, in dessen Stromazellen in den Abbildungen Ilbergs (1968 b) zahlreiche Mitochondrien gezeigt werden, spricht für eine gewisse Stoffwechselaktivität in diesem Bereich. Da hier eine hohe Resorptionsrate von Thorotrast beobachtet wurde, wird die Resorption von Perilymphe in diesem Bereich für möglich gehalten (Ilberg, 1968 b).

Die massiven SDH Aktivitäten in den inneren und äußeren Haarzellen sind bekannt und sprechen für ihre intensive metabolische Aktivität. Sie wurden in etwa der gleichen intrazellulären Verteilung bereits bei Vosteen (1956 a c) beschrieben. Die Stützzenen der äußeren Haarzellen jedoch, denen vielfach ebenfalls eine erhöhte Stoffwechselaktivität zugeschrieben wird (Spoendlin & Balogh, 1963 a, b 1964), zeigten in unseren Präparaten le-

diglich am Zellrand unterhalb der Basis der äußeren Haarzellen perlschnurartig aneinandergerahnte Farbstoffpunkte. Da dies recht genau der Lage der völlig von Plasmalemma und Cytoplasma der Deiterzellen umgebenen äußeren Spiralfasern entspricht (Spoendlin & Balogh, 1963 a b) muß man annehmen daß es sich hier um eine Darstellung von SDH in diesen Nervenfasern handelt. Die Deiterzellen selbst weisen demnach keinerlei Farbstoffablagerungen auf was den Befunden von Spoendlin & Balogh (1963 a b 1964) Gehrhardt (1961 a b) und Nakai & Hilding (1968) widerspricht. Auch die inneren Stützzellen zeigen keine Formazandepots.

In den Nervenendigungen an der Basis der äußeren und inneren Haarzellen fanden wir intensive Färbung in Übereinstimmung mit Vosteen (1960) und im Gegensatz zu Gehrhardt (1961 a b) und Spoendlin & Balogh (1963 a b 1964).

Wie in den Untersuchungen Spoendlins & Baloghs (1963 a b 1964) wurden auch bei uns im Bereich des Ligamentum spirale oberhalb des Ansatzes der Reißner Membran deutliche Aktivitäten gefunden. Takahashi & Kimura (1970) konnten bei elektronenmikroskopischen Arbeiten am Rhesusaffen in dieser Zone

mitochondrienhaltige Fibrocyten demonstrieren wiesen aber daraufhin, daß der Reichtum dieser Zellen an Organellen bei kleinen Tieren geringer ist, was sich mit den Beobachtungen Spoendlins & Baloghs (1963 a b 1964) deckt. Letztere schreiben diesem Bereich wichtige Funktionen bei der Perilymphresorption oder -sekretion zu.

Die Färbung von Teilen des Lig. spirale nach Formolpräinkubation und substratfreier Inkubation nach Substanzauswaschung mit Ausnahme der Gegend hinter der Prominentia spiralis und der der Scala vestibuli benachbarten Bereiche weist auf einen gewissen unspezifischen Charakter hin. Die übrigen positiven Strukturen scheinen nach Auswertung der Kontrollversuche spezifische Aktivitäten nachzuweisen.

Vosteen (1958) fand nach Dauerschallein-

wirkung mit einem Sinuston von 2 000 Hz 70 dB nach 4-5 Tagen und bei 85 dB nach 2 Tagen Veränderungen in den äußeren Haarzellen die auf einen bestimmten Bezirk der Basalmembran beschränkt waren. Die Schädigungen ließen sich schematisch in mehrere Stufen gliedern wobei eine Skala des Aktivitätsverlusts in den Nervenendigungen über stärkere Abnahme der Färbung der Zellen selbst bis zum völligen Aktivitätsverlust und histologischer Schädigung durchgezogen wird.

Rein histochemisch korrelieren unsere Befunde mit diesen Ergebnissen gut. Die fehlende enge Lokalisation der Schäden, auf die Windungen bezogen läßt sich für unsere Präparate durch das verwendete Breitbandgeräusch erklären.

Unser Beschallungsmodus wurde entsprechend den Verhältnissen am Lärmarbeitsplatz eingerichtet. Er unterscheidet sich von dem Vosteens erheblich dadurch, daß wir zwischen den einzelnen Belastungsphasen 15 Stunden lange Pausen einlegten. Wir finden demnach bei wesentlich längerer stärkerer Beschallung im Verhältnis zu den Versuchen Vosteens keine sicheren histologischen Veränderungen im Sinne von lichtmikroskopisch faßbaren Schädigungen des Zellkörpers. Daraus ist abzuleiten, daß selbst nach stundenlangem stärkeren Beanspruchung der Phonorezeptoren durch eine Lärmpause eine Erholung etwa im Sinne der Auffüllung eines Stoffwechseldepots möglich ist. Die erhebliche Bedeutung der Erholungsphase bei Dauerlärmeinwirkung wird dadurch unterstrichen. In unseren Präparaten finden wir jedoch einen deutlichen Aktivitätsabfall auch in den inneren Haarzellen die bei Vosteen frei von Veränderungen blieben. Dies scheint der erstmals von Spoendlin & Balogh (1963 a, b 1964) geäußerten Ansicht zuzustimmen daß die inneren Haarzellen eine geringere Stoffwechselreserve als die äußeren besitzen und daher bei der in unseren Versuchen insgesamt verlängerten Belastungsdauer doch eine Stoffwechselschädigung zeigen die nicht in gleichem Maße wie die äußeren Haar-

zellen Energiereserven durch Stoffwechselnebenwege mobilisieren können.

Feinstrukturelle Untersuchungen Spoendlin's (1958, 1970, 1971) zeigen bei kurzzeitiger Belastung mit 100–130 dB „weißem Rauschen“ typische Veränderungen der Mitochondrien zuerst in den Nervenendigungen und später in den Haarzellen mit Auftreten von zahlreichen osmophilen Einschlüssen in den Lysosomen der Zona intermedia als Ausdruck eines degenerativen Abbauprozesses. Wegen der mitochondrialen Bindung der SDH könnten derartigen Veränderungen mit Verminderung der SDH-Aktivität bereits einhergehen.

Herrn Prof. Dr. med. habil. H. G. Dieroff, Oberarzt der Univ. HNO-Klinik Jena, wird für die Erstellung des Beschallungsprogramms herzlich gedankt.

SUMMARY

In the normal cochlea of the guinea pig the SDH has been demonstrated by a perfusion method. The enzyme activity was localized exactly as in the results after incubation of sections and small tissue samples with the notable exception of a negative reaction in the stria vascularis and in nerve cells of the spiral ganglion. The SDH activity is significantly reduced in the outer and inner hair cells and the nerve endings of animals which were exposed to white noise of 110 dB for 63 hours (9 h noise, 15 h rest, interval at Saturday and Sunday). The decrease of enzyme activity was not restricted to any particular area of the cochlear duct. The results are discussed.

LITERATUR

- Arnold, W. & Ilberg, Ch. v. 1971 Verbindungswege zwischen Liquor und Perilymphraum. *Arch. Ohr Nas Kehlkopfheilk.* 198: 247.
- Balogh, K. 1965 Histochemical localization of acid phosphatase activity in teeth after decalcification with EDTA. *J. Histochem. Cytochem.* 13: 303.
- Belanger, L. F. 1953 Autoradiographic detection of S^{35} in membrane of the inner ear of the rat. *Science* 118: 570.
- Dieroff, H. G. 1971 Persönliche Mitteilung.
- Dieroff, H. G. & Beck, Chl. 1966 Experimentell-mikroskopische Studie zur Frage der Lokalisation der Lactatdehydrogenase im Innenohr und des später resultierenden bleibenden Hörschadens. *Arch. Ohr Nas Kehlkopfheilk.* 185: 1.
- Gehrbardt, H. J. 1961 a. Zur Methodik der Succinatdehydrogenasenachweis in der Meerschweinchen-schnecke. *Arch. Ohr Nas Kehlkopfheilk.* 178: 158.
- 1961 b. Zur Verteilung der Succinatdehydrogenase in der Meerschweinchen-schnecke. *Arch. Ohr Nas Kehlkopfheilk.* 179: 195.
- Geyer, G. 1969 Ultrahistochemie. Gustav Fischer Jena.
- Halbhuber, K. J. & Geyer, G. 1966. Über die Verwendbarkeit von Polyäthylenglykol 600 als Einbettungsmedium in der Gefriermikrotechnik mit besonderer Berücksichtigung von histochemischen Ferment und Lipidnachweisen. *Anat. Anz.* 119: 128.
- Ilberg, Ch. v. 1968 a. Elektronenmikroskopische Untersuchung über Diffusion und Resorption von Thoriumdioxid an der Meerschweinchen-schnecke. I. Mitteilung, Ligamentum spirale und Stria vascularis. *Arch. Ohr Nas Kehlkopfheilk.* 190: 415.
- 1968 b. III. Mitteilung, Labyrinth spiralis. *Arch. Ohr Nas Kehlkopfheilk.* 192: 163.
- 1968 c. IV. Mitteilung, Basilarmembran und Cortisches Organ. *Arch. Ohr Nas Kehlkopfheilk.* 192: 384.
- Iurato, S. 1961 Submicroscopic structure of the membranous labyrinth. II. The epithelium of Corti's organ. *Z. Zellforsch.* 53: 259.
- 1962. Ibid. III. The supporting structure of Corti's organ. *Z. Zellforsch.* 56: 40.
- 1967 Submicroscopic structure of the inner ear. Pergamon Press, New York.
- Kohde, Y., Yoshikawa, Y. & Morimoto, M. 1962. Some oxidizing enzymes in the cochlea. *Ann. Otol.* 71: 96.
- Lien, D. J. 1970 a. Ultrastructural localization of DPNH-Diaphorase in the cochlea. *Acta Otolaryng. (Stockh.)* 69: 32.
- 1970 b. Morphological and function of the interdental cell. *J. Laryng.* 84: 1241.
- Nakai, Y. 1965 Histochemical study of the stria vascularis in the inner ear by electron microscopy. *Ann. Otol.* 74: 326.
- Nakai, Y. & Hilding, B. 1968. Oxidative enzymes in the cochlea. *Acta Otolaryng. (Stockh.)* 65: 459.
- Ogawa, K. & Barnett, R. J. 1965 Electron cytochemical studies of succinic dehydrogenase and dihydronicotinamide-adenine dinucleotide diaphorase activities. *J. Ultrastruct. Res.* 12: 488.
- Schätzle, W. 1971 Histochemie des Innenohres. Urban & Schwarzenberg, München-Berlin-Wien.
- Schätzle, W. & Mösebeck, K. 1962. Histochemische Untersuchungen zur Blockierung der Succinatdehydrogenase in der Meerschweinchen-schnecke. *Arch. Ohr Nas Kehlkopfheilk.* 181: 49.
- Shenita, T. R. & Bowser, G. H. 1963 The perineural epithelium: nature and significance. *Nature* 199: 577.
- Spoendlin, H. 1958. Submikroskopische Veränderungen am Corti-Organ des Meerschweinchen nach akustischer Belastung. *Pract. Otorhinolaryng. (Basel)* 20: 197.
- 1966. The organization of the cochlear receptor. S. Karger, Basel.
- 1970. Auditory vestibular olfactory and gustatory

- organs. In *Ultrastructure of the peripheral nervous system and sense organs* (ed. A. Bischoff). Georg Thieme, Stuttgart.
- 1971 Primary structural changes in the organ of Corti after acoustic overstimulation. *Acta Otolaryng* (Stockh.) 71 166.
- Spoendlin, H. & Balogh, K. 1963 a. Histochemical localization of dehydrogenases in the cochlea of living animals. *Laryngoscope* 73 1061
- 1963 b Histochemische Darstellung von Dehydrogenasen in der Schnecke lebender Tiere. *Arch Ohr Nas Kehlkopfheilk* 182 579
- 1964 Licht und elektronenmikroskopische Darstellung von Dehydrogenasen in der Schnecke nach intratympanaler Injektion. *Pract Otorhinolaryng* (Basel) 26 159
- Takahashi, T. & Kimura, R. S. 1970. The ultrastructure of the spiral ligament in the Rhesus monkey. *Acta Otolaryng* (Stockh.) 69 46.
- Vosgien, K. H. 1956 a Die Darstellung der Bernsteinsäuredehydrogenase in der Schnecke des Meerschweinchens. *Arch Ohr Nas Kehlkopfheilk* 168 295
- 1956 b Die Tetrazolreaktion in der Meerschweinischnecke nach Blockierung der Zellatmung durch Kaliumcyanid. *Arch Ohr Nas Kehlkopfheilk* 169 415
- 1956 c Untersuchungen über den Grundstoffwechsel im Innenohr des Meerschweinchens. *Z Laryng Rhinol Otol* 35 400.
- 1958. Die Erschöpfung der Phonorezeptoren nach funktioneller Belastung. *Arch Ohr Nas Kehlkopfheilk* 172 489
- 1960. The histochemistry of the enzymes of the oxygen metabolism in the inner ear. *Laryngoscope* 70 351
- 1961 Neue Aspekte zur Biologie und Pathologie des Innenohres. *Arch Ohr Nas Kehlkopfheilk* 178 1

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DENSITY OF MUCOUS GLANDS IN A BIOPSY MATERIAL OF CHRONIC SECRETORY OTITIS MEDIA

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(Received April 10, 1972)

Abstract Biopsies were taken from the anterior part of the promontory in the course of tubulation through a paracentesis opening in 85 ears affected with chronic secretory otitis media in the secretory stage from 54 patients, mostly children. The specimens were stained by the PAS-alcian blue whole-mount method, and on each biopsy the number of glands was counted, their area measured directly and their density calculated. Glands were found in all ears but two from which the specimens were very small. The density varied within wide limits, from 1 to 31 glands per mm². In most ears the density was 3-10 glands per mm². The mean density in the entire material was 6.7 glands per mm², gland count per biopsy 7.5 in all, 743 glands and 105 mm² mucosa were investigated. 92.7% of the glands were active, 5.7% were transitional types, and only 1.6% were degenerated. In cases with large quantities of mucus and thickened mucosa the density as well as the number of glands per biopsy were greatest. There was a relationship between the severity of the secretory otitis in the secretory stage and the density of mucous glands. This further stresses that the mucous secretion which accumulates in the middle ear is an active product of the mucous glands.

Research into the mucous elements in the Eustachian tube and middle ear has been greatly intensified during recent years, not least because of their significance in understanding the pathology and pathogenesis of chronic secretory otitis media.

Studying biopsies from 40 ears affected with secretory otitis media, removed from the mucosa inferiorly and anteriorly to the round window Bendick (1963) found glands in 22% of the cases. Sadé (1966) found glands in biopsies of the promontorial mucosa in all his

20 cases of secretory otitis, whereas Gunder sen (1971), studying 189 biopsies removed anteriorly to the round window found no glands in any case. Paparella & Lim (1967) reported on gland-like structures in biopsies from the mastoid process in three patients with secretory otitis.

In autopsy materials of secretory otitis Sadé (1966) observed glands in the middle ear in three cases and de Moura & Hayden (1969) in one. All these studies (except Gundersen's) were based on histological sections. Previous biopsy as well as autopsy materials have not been subjected to determination of the density of glands or differentiation of the various types or secretory stages of glands.

For the study of mucous elements in the middle ear we have used whole-mount methods, preparing the mucosa in toto, counting all glands, making direct measurements of the mucosal area, and calculating the glandular density. In 1 patient with mild secretory otitis media we had found (Bak Pedersen & Tos, 1971) by post-mortem study of 370 mm² mucous membrane from the osseous Eustachian tube and middle ear 488 glands.

The glands were in different secretory stages (Tos & Bak-Pedersen, 1972): 1) Active glands with narrow ducts without stagnation of mucus and with well-preserved secretory epithelium. 2) Transitional varieties with dilated ducts and stagnant mucus, loss of the

secretory epithelium which however was preserved in places. 3) Degenerate, inactive glands, with pronounced cystic dilatation of the ducts and total loss of the secretory epithelium. On this basis, we advanced a new secretory pathogenesis of secretory otitis: Aetiological factors give rise to metaplasia of the middle-ear mucosa with formation of mucous glands (developmental stage) which condition lasts for several months. When the formation of the glands is completed, they secrete mucus (secretory stage) even though the aetiological factor which has caused their formation has ceased to exist. When most glands have started to degenerate the production of mucus decreases, and the disease improves spontaneously (degenerative stage), provided that it has not caused irreparable damage i.e. adhesive otitis.

Foetuses and newborn infants have no mucous glands in the middle ear or osseous Eustachian tube (Buch & Balslev Jørgensen, 1964; Tos, 1970, 1971). By studying almost the entire mucosa from the osseous tube and middle ear from 45 clinically normal adult temporal bones, Bak-Pedersen & Tos (1972) found that 41 (91%) had glands in the mucosa. However these glands were as a rule present in small numbers, less than 20 in 45%, more than 40 in 65% and less than 60 in 85% of the cases. Most of the glands were degenerate. The mean density within the entire material was 0.53 gland per mm². Thus, although there may be glands in clinically normal middle ears, they are probably not a normal component of the mucosa, but have arisen because of previous pathological actions upon the mucosa.

MATERIAL AND METHOD

The material comprises 85 ears from 54 patients treated by tubulation for chronic secretory otitis media. One patient was 2 years, 16 were 3-5 years, 26 were 6-10 years and 8 were 11-16 years of age. Three were adults. Under general anaesthesia, after paracentesis

and suction of mucus through the paracentesis opening, a biopsy specimen of the mucosa was taken anteriorly on the promontory at the junction to the anterior part of the hypotympanum. In about three-quarters of all cases one piece of mucosa was removed, in the remaining cases two or more small pieces. It was endeavoured to make the biopsy comprise the full thickness of the mucous membrane. After fixation in formal alcohol and staining by the PAS-alcian blue whole mount method the preparations were cleared in anise oil, microdissected, and smoothed under the stereomicroscope, placed in a chamber filled with anise oil-colophonium, and sealed with paraffin. In each biopsy all glands were counted in the stereomicroscope, magnification 40 \times ; the area of the biopsy was measured directly and the density was calculated. The number of active and of degenerate glands as well as of the transitional stages were determined separately for each biopsy.

RESULTS

All glands were tubular the majority consisting of one tubule running from the orifice on the epithelial surface either vertically or obliquely down into the mucosa (Fig. 1). Besides this, there were several branched glands, consisting of one excretory duct into which two tubules debouched. More rarely there were larger glands having several tubules. The various types of glands, their size and site, have been described previously in an autopsy specimen (Bak-Pedersen & Tos, 1971). In the present biopsy material we found the same types and approximately the same size of glands.

On the whole, the density of glands was high, but there were marked variations (Table 1). Only 3 cases were found to contain no glands. In most of the ears the density was 3-10 glands per mm² but in others a very high density was observed. The maximum density was 31 glands per mm². For all 85

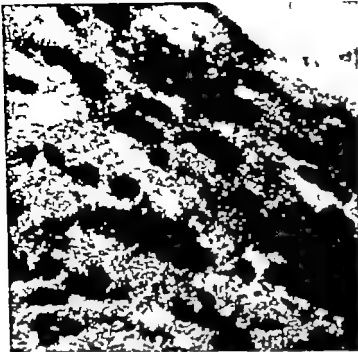


Fig. 1. A biopsy showing a great density of small tubular active glands the orifices of which are visible. PAS-alcian blue-stained whole mount, 200.

ears the mean density was 6.7 glands per mm^2 the mean area of the biopsy 1.3 mm^2 and the mean number of glands per biopsy 8.7. In all, 743 glands were found within the entire material and 105.3 mm^2 of mucosa were investigated.

The size of the biopsy influences the reliability of the density determination. In the 2 cases in which no glands were found, the mean area was small (Table I). The area of the smallest biopsy was 0.2 and that of the largest 6.7 mm^2 (Table II). In the group of small biopsies the density might vary widely. Within the group in which the area of the biopsy was 0.2–0.4 mm^2 the highest density was 31 glands per mm^2 but there was also one preparation with no glands. Within the group in which the biopsy area was 0.5–0.9 mm^2 there was also a marked variation in density and in the number of glands per biopsy. However a biopsy which is 0.5 mm^2 can afford a fairly reliable picture of the glandular density the mean density in this group being 5.7 glands per mm^2 . In most ears the biopsy was 1 mm^2 or larger. The reliability

of the density determination was considerably increased in biopsies of this size in particular the mean gland count per biopsy was large.

The highest gland count, in a biopsy of 3.8 mm^2 was 91 glands, corresponding to a density of 24 glands per mm^2 (Table II). In 20 ears the gland count per biopsy exceeded 10 in 5 ears it exceeded 30. The mean number of glands per biopsy in cases where the area was 3 mm^2 or over was very large.

Out of a total of 743 glands 689 (92.7%) were active, the entire tubule being lined with well-preserved secretory epithelium, with no dilatation of the duct and no stagnation of mucus in the duct (Fig. 1). 42 glands (5.7%) were of the transitional type, with incipient stagnation of mucus and incipient dilatation of the duct system, but having fairly well preserved secretory epithelium. Only 12 glands (1.6%) were completely degenerated, cystic, with dilated ducts, filled with stagnant mucus, and showing total disappearance of the secretory epithelium. The transitional and degenerate glands were found especially in biopsies

Table I. Number of ears in each density group the corresponding mean density area of biopsy and number of glands per biopsy

Density group (glands/mm ²)	No. of ears	Mean density	Mean area (mm ²)	Mean number of glands
0	2	0	0.6	0
1-2	16	1.8	1.0	1.6
3-5	29	3.8	1.3	5.4
6-10	23	7.6	1.3	9.5
11-15	9	13.1	1.5	19.8
16-20	4	17.5	1.0	17.2
21-40	2	27.5	2.0	48.0
Total	85	6.7	1.3	8.7

in which the gland count was high and which also contained many active glands. The very high percentage of active glands indicates that the disease has been in the secretory stage and fully explains the presence of mucus in the middle ear.

By tubulation the amount of mucous discharge could be determined (Table III). In cases with little or moderate discharge the mean density and mean number of glands per biopsy were lower than in cases where the middle ear was filled with mucus. In the group with but little secretion there were several cases in which the amount of secretion was minimal, in a few merely in the form of film of mucus on the mucosa. All ears with a little mucus were found to contain glands. Within the group with moderate quantities of secretion only one case did not exhibit glands.

Table II. Relationship between size of biopsy and variation in density as well as variation in number of glands per biopsy

Size of biopsy (mm ²)	No. of ears	Density			Number of glands per biopsy		
		Max.	Min.	Mean	Max.	Min.	Mean
0.2-0.4	6	31	0	10.5	5	0	4.3
0.5-0.9	27	18	0	5.7	11	0	3.4
1-1.9	36	18	1	6.3	34	1	7.8
2-2.9	11	13	1	5.3	32		11
3-3.9	3	4	4	12.3	91	1	43.3
5-6.7	2	14	5	9.5	68	34	5.0

Similarly there was one case without glands in the group with ample secretion. If the biopsy had been larger or if it had been taken from another site, it would no doubt have shown glands in these two cases too. Thus, there was a relationship between the quantity of secretion and the density of glands and thus between the severity of secretory otitis in the secretory stage and the gland density.

The thickness of the mucosa was assessed during the tubulation (Table IV). In cases where the mucosa was not thickened the density of glands as well as the number of glands per biopsy were lowest. The lower average number of glands per biopsy than the average density indicates that the biopsies were small, as is comprehensible since it is difficult to take a biopsy from a thin mucosa. Within this group there was one ear in which no glands were found. In cases having a slightly or moderately thickened mucosa the density as well as number of glands per biopsy were high, even higher than in cases with a greatly thickened mucosa among which there was also one ear without glands. In the group with a greatly thickened mucosa the mean density as well as the mean number of glands per biopsy were surprisingly low. A possible explanation is that they had oedema which caused the thickening of the mucosa. In cases with highly proliferating, almost polypous granulating mucosa, where small round polyps were removed from the mucosal surface at the time of taking the biopsy and could then be examined *in toto* there was a fairly large mean number of glands per biopsy (Table IV). Often, but not invariably, there were small young, active glands in the polyp which was covered with secretory epithellum, indicating marked proliferation of the mucosa and metaplastic new formation of glands and goblet cells. However there were also cases of polypous mucosa whose surface was only partially covered with goblet cells and in which glands were not yet found. Possibly such polyps had arisen recently and the formation of glands had not yet started.

DISCUSSION AND CONCLUSION

On the basis of the secretory pathogenesis Tos & Bak Pedersen (1972) have categorized secretory otitis into the developmental stage, the secretory stage, and the degenerative stage. As mucus was found in all the ears, in varying quantity it is true, the disease must have been in the secretory stage at the time that the biopsies were taken. The density of glands was relatively great. In 92.7% of the cases the glands were active and producing mucus. There is little doubt that the mucus which accumulates in the middle ear is an active product of these glands and of the goblet cells. The density proved to be widely varied, from 1 to 31 glands per mm² and this was due only partially to a relatively small biopsy. True, it was endeavoured to remove the biopsy from the same site in all cases, but the density need not be the same in all ears in the same area. In the autopsy specimen described previously the density varied very much between the different parts of the tympanic cavity being greatest in the hypotympanum anteriorly (Bak Pedersen & Tos, 1971), but this does not prove that the density is greatest at this site in all cases of secretory otitis. The main cause of the variations in density must be differences in the severity of the disease, which during the secretory stage depends upon the secretory capacity of the glands (their number and size), upon the secretory activity of the active glands, upon tubal function, and upon ciliary function which conveys the produced mucus to the

Table III *Relationship between quantity of secretion during tubulation and mean density of glands as well as mean number of glands per biopsy*

Quantity of secretion	No. of ears	Mean density of glands/mm ²	Mean number of glands per biopsy
Small	25	5.2	5.8
Moderate	13	5.5	5.8
	47	7.6	10.9

Table IV *Relationship between thickness of mucosa during tubulation and mean density of glands as well as mean number of glands per biopsy*

Thickness of mucosa	No. of ears	Mean density of glands/mm ²	Mean number of glands per biopsy
Not thickened	7	3.0	2.3
Slightly thickened	37	6.7	7.9
Moderately thickened	17	8.5	12.6
Considerably thickened	15	5.6	5.9
Polypus, granulating	9	7.5	14.5

rhinopharynx. In the mildest cases, with a small number of glands and a good tube as well as ciliary function the condition will presumably never progress to an accumulation of mucus and clinical symptoms. In the most severe cases, with a great density of glands, the mucus production will continue for years, in spite of a properly functioning drainage, and this will require several tubulations. This is confirmed by the present demonstration of a greater mean density and considerably larger number of glands per biopsy in cases with much mucus in the middle ear and with major mucosal changes. Another important factor influencing the variation in density is the time of taking the biopsy. Metaplasia of the mucosa and the formation of glands must take place gradually presumably in the course of a long period. If a biopsy is taken at an early stage of the disease there will presumably still be little density of glands, but it may continue to increase. In several cases we found signs that the glands were young and very small, suggesting that gland formation had not yet culminated and ceased. The very low percentage of degenerated glands also indicates that all ears were in the secretory stage and that the degenerative stage had not yet started. In the present biopsy material there is still a theoretical possibility that the density may increase further.

The total number of glands in each middle ear may be calculated to a certain degree: If

the area of the middle-ear mucosa is 400 mm² and the mean density is five glands per mm² there will be 2 000 glands in the middle ear and this represents a quite appreciable secretory capacity. However, very marked individual variations may be expected in area as well as in total gland number.

The present results confirm and supplement earlier findings by Bendek (1963), Sadé (1966) and by Paparella & Lim (1967) who demonstrated glands in the mucosa in secretory otitis, but are at marked variance with Gundersen (1971) who did not observe glands in any of his cases. This difference must be due to a difference in technique. Gundersen took his biopsies just anterior to the niche of the round window and smeared the material on the slide. In other words, these were not histological sections. Therefore, the failure to demonstrate glands does not prove that none were present. The whole-mount method demonstrates its superiority in the study of the mucous elements of mucous membranes. By this method it is possible not only to ascertain the presence of the glands, but also to examine each uncut gland separately and in this way determine the shape, size, and secretory capacity of the glands, their number and density.

Beside the mucous glands there are in the epithelium goblet cells. Owing to epithelial metaplasia their density is increased in secretory otitis. In the great majority of our biopsies we found a great density of goblet cells, but a quantitative study of their density cannot be evaluated until determinations of the goblet cell density in the middle ears of normal children have been carried out. We are at present performing quantitative studies of the goblet-cell density in the middle ear under normal as well as under abnormal conditions.

ZUSAMMENFASSUNG

Bei 85 Ohren von 54 Patienten (vorwiegend Kindern) mit Otitis media chronica im Sekretionsstadium wurden bei der Tubulation durch die Paracentheseöffnung Schleimhautbiopsien vom vordersten Teil des Promontoriums entnommen, und nach PAS-Alz¹ bi-

methoden der Ganzpräparate gefärbt. An jeder Biopsie wurde die Anzahl der Drüsen bestimmt, die Fläche gemessen und die Dichte berechnet. Nur bei zwei Ohren, wo die Biopsien klein waren, wurden keine Drüsen gefunden aber bei allen anderen. Die Dichte variierte von 1 bis 31 Drüsen/mm². Bei den meisten Ohren wurde eine Dichte von 3 bis 10 Drüsen/mm² gefunden. Die durchschnittliche Dichte des ganzen Materials war 6.7 Drüsen/mm². Anzahl der Drüsen pro Biopsie 7.5. Im ganzen wurden 743 Drüsen und 105 mm² Schleimhaut untersucht. 92.7% der Drüsen waren aktive, 5.7% Übergangsformen und nur 1.6% degenerierte Drüsen. Bei den Fällen mit viel Schleim und dicker Schleimhaut war die Dichte der Drüsen und Anzahl der Drüsen pro Biopsie am größten. Es wurde ein Zusammenhang zwischen Schweregraden der sekretorischen Otitis im Sekretionsstadium und der Dichte von Drüsen festgestellt, was auch wieder beweist, dass der Schleim, der sich im Mittelohr bei der sekretorischen Otitis ansammelt, ein aktives Produkt der mukösen Drüsen ist.

REFERENCES

Bak-Pedersen, A. & Tos, M. 1971. The mucous glands in chronic secretory otitis media. *Acta Otolaryng* (Stockh.) 72: 14.
— 1972. Mucous glands in the middle ear and osseous Eustachian tube in adult persons. *Acta Otol.* In press.
Bendek, G. A. 1963. Histopathology of transitory secretory otitis media. *Arch Otolaryng* (Chic.) 78: 33.
Buch, N. H. & Balslev-Jørgensen, M. 1964. Eustachian tube and middle ear. *Arch Otolaryng* (Chic.) 79: 472.
de Moura, L. F. P. & Hayden, R. C. 1969. Pathology of secretory otitis. A temporal bone report. *Henry Ford Hosp Med Bull* 17: 25.
Gundersen, T. 1971. Secretory otitis media. *Nord Med* 65: 945.
Paparella, M. M. & Lin, D. J. 1967. Pathogenesis and pathology of the "idiopathic blue ear drum". *Arch Otolaryng* (Chic.) 85: 249.
Sadé, J. 1966. Pathology and pathogenesis of secretory otitis media. *Arch Otolaryng* (Chic.) 84: 297.
Tos, M. 1970. Development of mucous glands in the human Eustachian tube. *Acta Otolaryng* (Stockh.) 70: 340.
— 1971. Distribution of mucous glands in the fetal Eustachian tube. *Arch Klin Exp Ohr Nas Kieferheilk* 197: 295.
Tos, M. & Bak-Pedersen, A. 1972a. The pathogenesis of chronic secretory otitis media. *Arch Otolaryng* (Chic.) 95: 511.

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THE ROLE OF THE SURFACE TENSION AND THE CONTACT ANGLES OF PERILYMPH IN STAPES SURGERY

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(Received April 5 1972)

Abstract. Physical aspects of stapes surgery are discussed. In order to determine the wettability of possible materials for the prosthetic use the advancing contact angles of perilymph on vitalium and Teflon were measured and were found to be $(62^\circ \pm 3^\circ)$ and $(101 \pm 4^\circ)$ respectively. As far as this physical factor is involved, the better choice is Teflon. In order to facilitate further *in vitro* experiments the surface tension of perilymph samples of 76 otosclerotic ears was measured. The distribution showed a mean value of 50.6 dynes/cm with a standard deviation of 3.3 dynes/cm.

As mentioned earlier (Marquet, 1970) the surface tension of perilymph is a very important factor in the protection of the labyrinth during footplate surgery. After stapedectomy the surface tension of perilymph, together with the contact angle of perilymph on the prosthesis, seem to determine the shape of the reparative soft tissue which will obturate the trepanation hole (Marquet et al., 1972). In order to determine the best suitable prosthesis for the stapes a comprehensive study of these factors was undertaken.

Behaviour of Perilymph in Contact with Prosthesis

The final result of a surgical intervention depends greatly on the shape of the obturating, reparative tissue. When a grafting material is used, there is some risk of obtaining a muff. When no graft is placed, the same complica-

tion can occur more frequently if a non-suitable material is used for the prosthesis and if all directions as given in our previous paper are not followed carefully.

We have observed that the wettability of the prosthesis and thus the contact angle θ of perilymph on the material used is very important.

Let us first consider the case of $\theta < 90^\circ$ (Fig. 1a). The liquid will wet and cover the prosthesis and the tissue growing on the meniscus will form a muff. When a suitable material (with $\theta > 90^\circ$) is placed carefully (without pushing the prosthesis too far into the perilymph) a more favourable situation is created (Fig. 1b). As described in a previous paper the tissue will now grow underneath the piston and really obturate the opening.

Measurement of Contact Angles

A useful estimate of the wettability of solids by a fluid is the advancing contact angle θ_a . This is the maximum value of θ when a liquid advances over a solid surface, while the receding angle θ_r is the minimum value when a liquid recedes over a solid surface (Johnson & Dettre, 1969). The advancing angle can be obtained by increasing the volume of a liquid drop resting on a plane solid surface, until the three-phase boundary moves over the sur-

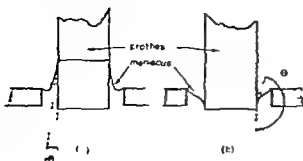


Fig 1 Shape of the perilymph meniscus: (a) contact angle θ of the perilymph on the prosthesis $< 90^\circ$; (b) $\theta > 90^\circ$.

face (drop method). Values of θ of perilymph on vitallium and polytetra fluoroethylene (PTFE or Teflon) were measured directly on the profile of the drop. The micro syringe which was used to increase the drop volume was kept immersed in the drop during the measurement (see Fig. 2).

As found by Bartell & Björklund (1952) the presence of this fine tip within the drop does not alter the value of the contact angle. The direct measurement of θ was done using a binocular loupe fitted with one goniometer eyepiece.

This technique was chosen because it can be used on solids having small surface area and requires only small amounts of liquid (less than $1 \mu\text{l}$). Contact angles were measured with an instrumental accuracy of

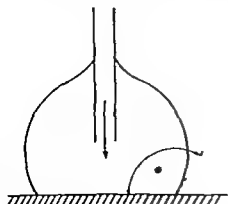


Fig 2 Technique for obtaining advancing contact angles.

about 2%. Samples from 4 patients were analyzed. For the advancing contact angle of perilymph on vitallium we found a mean value (θ) = 62° with a standard deviation of 3 and for perilymph on Teflon (θ) = 101° with a standard deviation of 4. In both cases the receding angles were very small ($< 15^\circ$) but could not be determined accurately because of the difficulties in locating the point of contact and in constructing the tangent to the drop profile in the contact point.

From these measurements we can conclude that from a physical point of view Teflon is the better suited material.

Determination of Surface Tension of Perilymph

The surface tension of perilymph accounts for the protection of the labyrinth against waste particles with a density greater than that of perilymph. The knowledge of the value of the surface tension of perilymph is also required to enable further "in vitro" experiments. For this purpose a microtensiometer was developed (Creten et al., 1972) enabling quick routine determination of the surface tension from samples of 0.4 to $0.7 \mu\text{l}$ with an accuracy of about 2%.

During footplate surgery the perilymph was obtained, after the footplate was perforated by means of a capillar with an outer diameter of about 0.6 mm and an inner one of about 0.3 mm. From each patient an amount of 0.5 to $2.0 \mu\text{l}$ was taken in one or more capillars. This amount was transferred into a micro syringe and a known quantity (0.4 to $0.7 \mu\text{l}$) was put in the glass receptacle of the microtensiometer. All manipulations including the determination of the surface tension of the sample were performed within a few minutes. The results obtained with samples from 76 patients are given in Fig. 3. All measurements were performed at 22°C .

The distribution has a mean value of 406 dynes/cm with a standard deviation of 33

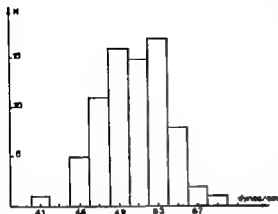


Fig 3 Distribution of the surface tension of perilymph.

dynes/cm. We may assume that the distribution is gaussian as the skewness g_1 and the kurtosis g_2 (Snedecor 1956) are both zero within the statistical errors.

$$(g_1 = -0.26 \pm 0.28, g_2 = 0.1 \pm 0.5)$$

As the patients were under a general anaesthesia called "protected sleep" (Delaruelle & Marquet, 1971) a surgical field with minimal bleeding was obtained, so that the perilymph samples were seldom polluted with blood. From 8 patients more than one sample was taken, one of which was polluted with blood. No detectable alteration of the surface tension of perilymph is found.

RÉSUMÉ

Ce travail se rapporte à l'étude des aspects physiques de la chirurgie de l'étrier. Afin de déterminer le degré de mouillage de différents types de prothèses, l'angle de contact de la périlymphe a été mesuré respectivement sur du vitallium et du téflon. Les résultats furent ($62^\circ \pm 3^\circ$) et ($101^\circ \pm 4^\circ$). Pour autant que ce paramètre physique soit déterminant, préférence est donnée au téflon. Afin de permettre une expérimentation ultérieure (in vitro), la tension superficielle de la périlymphe fut mesurée chez 76 patients

opérés d'otosclérose. La distribution donna une valeur moyenne de 50,6 dynes/cm avec une déviation standard de 3,3 dynes/cm.

ZUSAMMENFASSUNG

Die physikalischen Aspekte der Stapeschirurgie wurden besprochen. Um den Befechungsgrad des eventuellen Prothesenmaterials festzustellen, wurde der fortschreitende Kontaktwinkel von Perilymphe auf Vitallium und Teflon gemessen. Die respektiven Ergebnisse waren $62^\circ \pm 3$ und $101^\circ \pm 4$. Soweit dieser physikalische Parameter ausschlaggebend ist, ist Teflon zu bevorzugen. Um weitere in-vitro-Versuche zu ermöglichen, wurde die Oberflächenspannung von Perilymphe-Proben von 76 Otosklerose-Patienten gemessen. Die Verteilung ergab einen mittleren Wert von 50,6 dynes/cm mit einer Standardabweichung von 3,3 dynes/cm.

REFERENCES

- Bartell, F. E. & Björklund, C. W. 1952. Hysteresis of contact angles. *J Phys Chem* 56 453.
- Creten, W. L., Van Camp, K. J. & DeGraeme, W. F. 1972. A microtensiometer. *Phys Med Biol* 17 299.
- Delaruelle, J. & Marquet, J. 1971. A technique of anaesthesia, especially adapted for ear surgery "Protected Sleep". *Acta Otolaryng (Stockh.)*, Suppl. 281.
- Johnson, R. E. & Dettre, R. H. 1969. *Surface and colloid science* (ed. E. Matijevic) vol. 2, pp. 85-153. Wiley-Interscience, New York.
- Marquet, J. 1970. Causation and prevention of sensorineural hearing loss after ear surgery. *Ciba Foundation Symposium on Sensorineural Hearing Loss* (ed. G. E. W. Wolstenholme and J. Knight), pp. 313-323. J. and A. Churchill, London.
- Marquet, J., Creten, W. L. & Van Camp, K. J. 1972. Considerations about the surgical approach to stapedectomy. *Acta Otolaryng (Stockh.)* 74 406.
- Snedecor G. W. 1956. *Statistical methods*. Iowa State University Press, Ames, Iowa.

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TRANSTRACHEAL CERVICAL APPROACH FOR OPERATING ON SEVERE INTRATHORACIC TRACHEAL STENOSIS FOLLOWING ASSISTED RESPIRATION

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Abstract This report describes the surgical treatment of 5 patients with severe degrees of intrathoracic tracheal stenosis following long-term assisted respiration. The operation was carried out via a transtracheal approach in the cervical part of the trachea. Using long-handled instruments and local anaesthesia, the circular stenosed area was extirpated endotracheally. A cannular prosthesis was inserted and left in position for 4 months. The operations involved no complications, and the results were excellent in all five cases. The effect of surgery was evaluated by spirometry and body plethysmography which were performed before and 6 to 78 months after removal of the prosthesis.

The modern treatment of respiratory failure has saved patients' lives to an extent that could never have been imagined earlier. A serious complication following respirator therapy in resuscitation is stenosis in the intrathoracic part of the trachea.

Thorax surgeons have in these cases carried out a circular resection of the stenosed tracheal area and then sutured the trachea end to end (Michelson et al. 1961, Grillo 1965, 1969). This end-to-end method has brought the best results in the resectioning of a tracheal segment with an endotracheal tumour. The peritracheal region in these cases has been normal and permitted mobilization and suturing of both tracheal ends. When the same end-to-end operation is to be carried out in intrathoracic tracheal stenosis, several serious problems arise.

- 1 Peritracheal scar tissue and infection
- 2 The trachea is not easily mobilized for suturing
- 3 The sutures tend to "cut through"
- 4 Large blood vessels are often adherent to the trachea
- 5 Recurrent nerves, partly embedded in the scar tissue, are difficult to identify
- 6 Recurrent nerves can be injured in the postoperative cicatrization process.

Cases of sternal osteitis following sternal division, mediastinitis and rupture of the large blood vessels adherent to the trachea have been reported (Aboulker et al. 1967, Aboulker 1968) treated with the end-to-end technique. Even if just one recurrent nerve is injured intra- or postoperatively resulting in unilateral recurrent nerve paralysis, the consequences are not agreeable for the patient. Respiratory function tests have shown that unilateral recurrent nerve paralysis means that the tracheal lumen is reduced by one-third. Even a successful operation of the tracheal stenosis itself in many cases may bring no relief of the patient's dyspnoea if the result is eliminated by a unilateral recurrent nerve paralysis.

The published series of resection and end-to-end anastomosis, even when carried out by skilled hands, have shown a proportion of mortality (Pearson et al. 1968, Grillo 1969).



Fig. 1 Roentgenogram of intrathoracic tracheal stenosis.

MATERIAL AND METHOD

In the Department of Otorhinolaryngology University Central Hospital, Helsinki, we have developed a method for operating these severe cases of chronic intrathoracic tracheal stenosis (Fig. 1). Five have been treated to date. Table I shows the series and the results.

Owing to the numerous inconveniences of operating on the thoracic portion of the trachea in intubation anaesthesia (Nissen 1961 Woods et al. 1961) all the operations of the present series were performed under local anaesthesia. Preoperatively the patients were given atropin. We used 0.5% Carbocain® for infiltration even the stenosed area inside the trachea was infiltrated. The mucosal surfaces were anaesthetized with 3% Xylocain®. Diamorphine was used in doses of 5 mg at a time, in a total dose of 45 mg and Silomat® in doses of 20 mg at a time, up to a total of 80 mg per operation.

The patient lay on his back with a pillow under the shoulder blades, by which means the intrathoracic part of the trachea was slightly raised cranially. The skin incision was made vertically down to the jugulum. The surgeon then sat down behind the patient's head and made a vertical incision in the trachea, as distally as possible (Fig. 2). The edges of the tracheal incision were retracted. The stenosed ring inside the trachea was infiltrated by a long injection needle. The entire intratracheal stenosed area was then excised, using a knife with a long narrow handle and long handled scissors and tweezers (Fig. 3). The excision was continued until it reached tracheal lumen of a normal size. Sometimes the mediastinum was faintly visible through the thin tracheal wall. The next step was to insert a long and thick silver cannula encased in plastic tubing (Fig. 4). The distal end of the cannular prosthesis must reach below the excised cicatrized length. In some cases the distal end lay just above the carina. The incision was sutured,

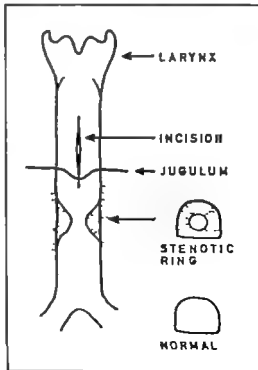


Fig. 2. Schematic appearance of the stenotic ring and the level of the incision.

curves (Poppius, 1969) and flow volume curves (Jordanoglou & Pride 1968)

All 5 patients had spirometrical evidence of airway obstruction during expiration as well as inspiration and flow-volume loops compatible with upper airway stenosis. All patients had normal values for transfer factor arterial blood gases, physiological right-left shunt and distribution tests based on analysis of expiratory CO₂ curves, suggesting absence of significant pulmonary parenchymal disease. One patient (case 2) with a clinical diagnosis of asthma and a history of persistent sputum showed a slight increase in both nitrogen single breath index and arterial-end-tidal CO₂ difference compatible with peripheral airways disease in the other 4 patients these tests gave values within the normal range.

Preoperatively spirometry showed that both expiratory and inspiratory flow were greatly increased in all patients, excepting the patient with asthma (case 2, Table II). Of the 4 patients examined with body plethysmography both before and after surgery all but case 2 showed a remarkable postoperative fall in airway resistance to normal or borderline values. In case 2, surgery was followed by no change in FEV₁, a marked increase in inspiratory flow (FIV%) a reduction of airway resistance by two-thirds to a still definitely pathological value and a change in the shape of the flow-volume curve from the type of upper airway stenosis to the type of peripheral airways obstruction (Jordanoglou & Pride 1968). In this case the upper airway stenosis had presumably prevented expiratory airway collapse before the operation, and surgery resulted in a partial shift of the site of obstruction to air flow from the area of the tracheal stenosis towards the periphery.

Analysis of respiratory function showed severe upper airway stenosis before operation in all 5 cases. In 4 cases, surgery re-established normal or almost normal flow conditions. In the asthmatic patient (case 2) the effect of surgery was somewhat difficult to evaluate owing to the variable peripheral airway ob-

Table II Results of spirometry and body plethysmography before and 6 to 28 months after removal of the prosthesis

Case no	Sex	Age (yrs)	Height (cm)	Parameter of respiratory function	Before surgery	After surgery
1	♀	23	172	VC	3.38	3.99
				FEV ₁	1.29	2.02
				FEV ₂₅	39	75
				FIV	42	90
				Ri	— ^b	4.2
2	♀	21	161	Ri TGVm	—	13.4
				VC	3.57	2.96
				FEV ₁	1.10	1.80
				FEV ₂₅	32	48
				FIV	36	71
3	♀	45	154	Ri	28.8	8.3
				Ri TGVm	131.4	42.4
				VC	2.92	2.85
				FEV ₁	1.39	1.25
				FEV ₂₅	49	42
4	♂	38	173	FIV	54	109
				Ri	17.4	1.4
				Ri TGVm	56.0	3.8
				VC	4.22	4.60
				FEV ₁	1.69	3.30
5	♀	20	164	FEV ₂₅	40	79
				FIV	34	86
				Ri	19.9	3.0
				Ri TGVm	94.2	10.5
				VC	3.74	4.19
				FEV ₁	1.56	3.00
				FEV ₂₅	48	91
				FIV	57	94
				Ri	5.9	1.3
				Ri TGVm	4.0	6.4

VC and FEV₁ in l BTPS. Ri in cm H₂O/l's Ri TGVm in cm H₂O s.

^b Body plethysmography not possible due to immobilization of fractured leg.

struction there was, however indirect physiological evidence of a marked reduction of the degree of upper airway stenosis also in this case.

DISCUSSION

Intubation anaesthesia could not be used because of the shortage of space in the trachea and in order to avoid the risk of the anaesthetic gases being forced into the mediastinum by overpressure.

The method presupposes the use of kr

handled instruments for the transtracheal intrathoracic operations. We realized there was a risk of acute haemorrhage in the operative field. Such a haemorrhage, if massive, would be difficult to control. In no single case, however did a profuse bleeding occur. This was probably because the operation was not performed while granulation was in progress, since we waited until cicatrization was complete and mature in the form of a totally fibrotized and almost avascular scar tissue. At the same time we applied anaesthesia so as to ensure the least possible irritation that might cause coughing. Silomat® and Diazepam® in large doses proved to be well-suited for this purpose.

The cannular prosthesis, on completion of the operation, was inserted into the trachea without in any way covering the surfaces of tracheal wounds caused by scar excision. During the 4 months the cannular prosthesis was in position the wound surfaces were completely epithelialized.

A straight prosthesis of the Aboulker type (Aboulker 1968) was used for 2 patients (cases 2 and 4). This prosthesis had been successfully used by the present author (Grahne 1971) in operations for subglottic laryngeal stenosis. In case 2, because of difficult respiration, the prosthesis had to be exchanged for a cannular prosthesis on the very day of the operation. In case 4 the exchange had to be effected only 3½ months after the operation since the distal end of the straight prosthesis had produced a new tracheal stenosis. A later re-operation of the new stenosis using the cannular prosthesis was successful. The prosthesis in the intrathoracic part of the trachea must necessarily follow the natural, slightly curved shape of the trachea, and the gently curved cannular prosthesis used in most cases of the present series suited the purpose admirably.

The results have been excellent. It is the experience of the surgical member of the team (B.G.) that the Aboulker prosthesis, which is first-rate in subglottic stenosis, cannot be used

intrathoracically because of the slight curve in this part of the trachea.

ZUSAMMENFASSUNG

Es wird die chirurgische Behandlung von fünf Patienten mit schwerer intrathorakaler Trachealstenose im Gefolge von langfristiger Respirator-Behandlung beschrieben. Die Operation wurde auf transtrachealem Wege im zervikalen Abschnitt der Trachea ausgeführt. In Lokalanästhesie wurde mit Hilfe von langschäftigen Instrumenten die narbige Ringstenose endotracheal excidiert. Es wurde eine kanülentprothese eingelegt und vier Monate lang liegen gelassen. Die Operationen verliefen ohne Komplikationen, und die Resultate waren in allen fünf Fällen ausgezeichnet. Der Erfolg der chirurgischen Behandlung wurde mit Spirometrie und Körperplethysmographie kontrolliert, die vor der Operation sowie 6-28 Monate nach Entfernung der Prothese vorgenommen wurden.

REFERENCES

- Aboulker P. 1968 Traitement des sténoses trachéales. *Probl. Actuels Otorhinolaryng.* p. 275.
- Aboulker P., Desmarest, J. E. & Sannou, G. 1967 Réflexions à propos de deux nouveaux cas de sténose trachéale traités par résection anastomose. *Ann. Otolaryng. (Par.)* 84: 771.
- Engstrom, H., Grimby G. & Söderholm, B. 1964 Dynamic spirometry in patients with tracheal stenosis. *Acta Med Scand* 176: 329.
- Grahne, B. 1971 Operative treatment of severe chronic traumatic laryngeal stenosis in infants up to three years old. *Acta Otolaryng. (Stockh.)* 72: 134.
- Gribo, H. C. 1965 Circumferential resection and reconstruction of the mediastinal and cervical trachea. *Ann. Surg.* 162: 374.
- 1969 The management of tracheal stenosis following assisted respiration. *J. Thorac. Cardio. Surg.* 57: 52.
- Jordanoglou, J. & Pride N. B. 1968 A comparison of maximum inspiratory and expiratory flow in health and lung disease. *Thorax* 23: 38.
- Michelson, E., Solomon, R., Mason, L. & Ramirez, J. 1961 Experiments in tracheal reconstruction. *J. Thor. Cardio. Surg.* 41: 74.
- Moyers, K., Smidt, U. & Buchheim, F. W. 1969 Verbesserung ganzkörperplethysmographischer Untersuchungen durch Einsatz eines Analogrechners. *Pflüger Arch. Ges. Physiol.* 307: 11.
- Niisea, R. 1961 Extracorporelle Zirkulation für langdauernde (30 Minuten) Atemunterbrechung zur Operation bifurkationsnaher Trachealgeschwülste. *Schweiz. Med. Woch.* 91: 957.
- Ogilvie, C. M., Fowler, R. E., Blackmore, W. S. & Morton, J. W. 1957 A standardized breath holding technique for the clinical measurement of the

- diffusing capacity of the lung for carbon monoxide. *J Clin Invest* 36 1
- Pearson, F. G., Goldberg, M. & Da Silva, A. J. 1968 Tracheal stenosis complicating tracheostomy with cuffed tubes. *Arch Surg (Chic.)* 97 380
- Poppius, H. 1969 Expiratory CO₂ curve in pulmonary diseases. *Scand J Resp Dis* 50 135
- Poppius, H., Varpila, E. & Korhonen, O. 1969 Respiratory function and exercise tolerance in relaxation of the diaphragm. *Scand J Resp Dis* 50 228
- Sandqvist, L. & Kjellmer, I. 1960. Normal values for the single breath nitrogen elimination test in different age groups. *Scand J Clin Lab Invest* 12 131
- Wohowitz, H. J., Buchheim, F. W. & Wohowitz, E. 1967 Zur Theorie und Praxis der Ganzkörper plethysmographie in der Lungenfunktionsanalyse. *Prax Pneumol* 21 449
- Woods, F. M., Neptune, W. B. & Palach, A. 1961 Resection of the carina and main stem bronch with the use of extracorporeal circulation. *New Eng J Med* 264 492.

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EFFECT ON CARIES SUSCEPTIBILITY AFTER SURGICAL TREATMENT OF DROOLING IN PATIENTS WITH NEUROLOGICAL DISORDERS

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(Received April 9 1972)

Abstract In 10 patients with neurological disorders and drooling, retroposition of the submandibular and ligation of the sublingual ducts was performed in order to reduce the drooling. This study shows the effect of the operation on the caries activity. Examination of the dental status was made before and one year after the operation. In some cases an enhanced susceptibility to caries was found, measured as increase in white spot lesions or transformation of these lesions into caries cavities. With good prophylactic measures the risk of increasing caries activity would not appear as a major contra-indication to the surgical treatment described.

Drooling can be seen in persons with neurological diseases, e.g. cerebral palsy. Drooling has been treated either with anticholinergic drugs or surgically to reduce saliva production (Goode & Smith, 1970; Enfors & Lundberg, 1968). Some authors have described operative methods to reduce the accumulation of saliva in the anterior part of the mouth. Laage-Hellman (1969) describes a surgical method for retropositioning of the submandibular ducts. The operation was performed on a few drooling children with cerebral palsy. A similar operation was described by Wilkie (1967) who in a few cases of drooling children made a retroposition of the parotid ducts. In a second stage, bilateral extirpation of the submandibular glands was performed.

A modification of the surgical technique described by Laage-Hellman (1969) was elaborated (Ekedahl, 1973) to control the drooling.

In this technique the duct from the submandibular gland was led to the palatal arches and the ducts from the sublingual glands were ligated.

Surgical treatment involving changes in salivary secretion may increase the susceptibility to oral disease since the composition and rate of secretion of saliva is of importance to counteract the caries provoking effects of food debris and bacteria-rich plaques retained on the teeth.

The present investigation was performed to study the effect of the operation on the caries activity. A preliminary report of these data has been presented (Ericson et al., 1971).

MATERIAL AND METHOD

The investigation was made on 5 male and 5 female patients, 7 to 26 years of age. Clinical and laboratory data were collected before and one year after operation.

Oral examination

Caries index, dental plaque index and gingival index were recorded. The dental status was checked with an explorer and mirror in a dentist's chair under good lighting conditions. No roentgenological examination was made.

Table I Caries plaque and gingival index of drooling persons before and after operation a.m. Ekedahl (1973)

		Caries Index						White spot lesions		Plaque Index		Gingival Index		
Age	Sex	DMFT		DMFS		Westin		Before	After	Before	After	Before	After	
		Before	After	Before	After	Before	After							
K. A.	10	♀	2	4	2	4	0.02	0.04	0	0	2.5	1.1	1	1
C. P.	11	♀	6	11	7	12	0.06	0.12	0	34	1.5	1.3	1	1.2
B. J.	18	♀	26	27	64	69	0.55	0.60	0	0	2.7	1.7	1.2	1
L. U.	18	♀	10	14	22	27	0.16	0.19	2	4	3	1.5	2	2
B. O.	19	♀	3	6	6	15	0.04	0.09	0	0	3	2.4	2.3	2
M. N.	7	♂	8	11	10	17	0.10	0.15	6	2	1.5	1.4	1	1
P. J.	12	♂	1	2	1	2	0	0.01	0	0	2.8	1.9	3	2
J. P.	1	♂	18	22	32	36	0.25	0.29	0	5	3	2.4	3	3
L. S.	21	♂	7	9	7	9	0.07	0.09	0	0	2	1	1.2	1.2
B. K.	26	♂	18	18	27	29	0.20	0.21	0	11	3	2.3	3	3

Caries index was scored according to the DMF system (Klein et al. 1938). In Table I DMFT stands for decayed, missing and filled teeth and DMFS for decayed, missing and filled surfaces. The maximal DMFT number is 28 and the maximal DMFS number is 128. The Westin index is calculated as the ratio of decayed and filled surfaces to 100 selected surfaces of 28 teeth. Missing incisors are evaluated as three surfaces and missing premolars or molars as four surfaces.

The plaque index (Silness & Loe 1964) describes the distribution of soft bacteria-rich material on the teeth.

Scores Criteria

- 0 No plaque
- 1 A film of plaque adhering to the free gingival margin and adjacent area of the tooth. The plaque may be seen in situ only after application of disclosing solution or by using the probe on the tooth surface
- 2 Moderate accumulation of soft deposits within the gingival pocket or on the tooth and gingival pocket, or on the tooth and gingival margin which can be seen with the naked eye
- 3 Abundance of soft matter within the gingival pocket and or on the tooth and gingival margin

The gingival index (Löe & Silness, 1963) describes the degree of inflammation of the gingiva.

Scores Criteria

- 0 Absence of inflammation.
- 1 Mild inflammation—slight change in colour and little change in texture
- 2 Moderate inflammation—moderate glazing redness, oedema and hypertrophy
Bleeding on pressure.
- 3 Severe inflammation—marked redness and hypertrophy
Tendency to spontaneous bleeding.
Ulceration.

Both plaque and gingival index can take a maximal value of 3.0 units.

White spot lesions are early caries lesions recognized as a white opacity in the cervical area of the enamel along the gingival margin. It is characterized microscopically as a subsurface lesion under an intact surface. They are an indication of high caries activities.

RESULTS

The results from the oral examination are shown in Table I. Relatively low values of car-

ries index are seen. The number of white spot lesions has increased after the operation. In one case (C. P.) 34 such lesions had developed and in another (B. K.) 11 lesions were found one year after operation. Also 5 new lesions (J. P.) indicate an increase in caries activity. In some persons (M. S. and L. U.) some of the white spot lesions noted at the first examination had at the later examination developed into caries cavities. Both before and after the operation large amounts of dental plaque and high values for gingival inflammation were observed.

DISCUSSION

The severe gingivitis of the children in this study corresponds well to the high plaque indices scored. The dental status is on the other hand remarkably good considering the poor oral hygiene. The presence of dental plaque to the extent found here is normally not found in persons of corresponding ages and still only 2 of the test persons (B. J. and M. S.) had caries indices higher than average in their respective age groups. B. J. age 18 thus had a DMFS index of 64 which can be compared with an index number of 40 reported by Anderson (1961) as the mean for 566 18-20-year-old men entering military service. M. S. 7 years old, had a DMFS index of 11 when the mean DMFS index of 1 275 children of this age in the Norrköping study was 5.15 (Sellman & Syrrist, 1968).

The surgical treatment thus might have turned the balance for those whose caries activity earlier was kept low probably mainly due to a diet low in carbohydrates. Three of the cases, C. P., J. P. and B. K. have developed a severe increase in early carious lesions in the form of white spot lesions. Such early lesions, which are found in cases with high caries activity and developed in the cervical regions on the buccal surfaces, are by convention not included in the caries index numbers and are therefore reported separately.

The diet of these institutionalized children is well balanced with regard to calories and nutrients. Fruit like apples, oranges, bananas and pineapples are served between meals and as deserts. Only occasionally—at nine dinners during a 5-week period—are sweetened desserts served. Sweets are given only on Saturdays. The composition of the diet and a probable but not measured, high rate of salivary flow may account for the relatively low caries frequency observed. The flow rates could not be determined due to the lack of capability of cooperation of the children.

Since no changes in diet and only minor changes in prescribed medicine had occurred during the test period, saliva seems to be the factor that can be correlated to the increased caries activity. The increase is seen as higher number of white spot lesions and change from the lesions into carious cavities. The composition of the salivary secretion is obviously changed due to the inactivation of the sublingual gland. The relative importance and influence of the individual secretions in various parts of the oral cavity is also altered after operation. The liquid environment for the incisors is perhaps most affected and in this region the new white spot lesions are found. The composition of the whole secretion has been studied but will be reported elsewhere. It can be mentioned, however that a decrease in buffer capacity values and an increase of the protein content were found in the test persons one year after operation.

The surgical treatment thus seemed to enhance the susceptibility to caries. A good effect of the operation on the drooling that these children suffer from, would improve the social adjustment for the children. An enhanced caries susceptibility would then not appear as a major contraindication to the surgical treatment. On the other hand it must not be neglected and full cooperation with dentists to make use of optimal caries preventive measures is recommended. The nursing personnel should be trained to assist in the caries prevention program.

ZUSAMMENFASSUNG

Zehn Patienten mit neurologischen Krankheiten und Gelfer wurden mit Retroposition der Ausführungsgänge des Gland Submandibularis und Ligatur der Ausführungsgänge des Gland. Sublingualis operiert, um den Gelfer zu vermindern. Vortliegende Arbeit beschreibt den Effekt der oben beschriebenen Operation auf die kariogene Aktivität. Untersuchungen des Zustandes der Zähne wurden vor und ein Jahr nach der Operation gemacht. Eine Zunahme der kariogenen Aktivität wurde in einigen Fällen als Vermehrung von oberflächlichem Schmelzkaries oder Übergang von diesem oberflächlichen Schmelzkaries zu Karies registriert. Mit guter oraler Hygiene soll die Gefahr einer Zunahme der kariogenen Aktivität nicht als Kontraindikation der chirurgischen Behandlung gelten.

REFERENCES

- Andersson, T. 1961 Investigation of caries prevalence in 18-year-old men in southern Sweden. *Odont Rev* 12 45.
- Ekedahl, C. 1973 Surgical treatment of drooling. *Acta Otolaryng* (Stockh.) In press.
- Enfors, B. & Lundberg, Å. 1968 Behandling av hyper-salivation hos CP-barn. *Svensk Läkertidn* 65 4416.
- Ericson, Th., Nordblom, A. & Ekedahl, C. 1971 Data on saliva from children suffering from cerebral palsy. *J Dentul Res* 50 1223.
- Goode, R. L. & Smith, R. A. 1970. The surgical management of sialorrhea. *Laryngoscope* 80 1078.
- Klein, H., Palmer, C. E. & Knutson, J. W. 1938 Dental status and dental needs of elementary school children. *US Publ Health Repts* 53 751.
- Laage Hellman, J. E. 1969 Retroposition av gl. submandibularis utförsgång som behandling vid drooling. *Nord Med* 82 1522.
- Loe, H. & Silness, J. 1963 Periodontal disease in pregnancy I. Prevalence and severity. *Acta Odont Scand* 21 533.
- Sellman, S. & Syrjäst, A. 1968 The Norrbörs fluoridation study. *Odont Rev* 19 23.
- Silness, J. & Loe, H. 1964 Periodontal disease in pregnancy II. Correlation between oral hygiene and periodontal conditions. *Acta Odont Scand* 22 121.
- Willie, T. F. 1967 The problem of drooling in cerebral palsy: A surgical approach. *Canad J Surg* 10 60.

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ACTA OTO-LARYNGOLOGICA

Vol. 75 No. 2-3

COMPTE RENDU
DE LA RÉUNION SCIENTIFIQUE DU

COLLEGIUM
OTO-RHINO-LARYNGOLOGICUM
AMICITIAE SACRUM

BERNE, LE 10-14 SEPTEMBRE, 1972

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NARVÄVÄGEN 16, 11523 STOCKHOLM



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President of the
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André Schroeder
Dean of the Faculty
of Medicine

Opening Session September 11 1972

INTRODUCTION

F Escher

Liebe Mitglieder des Collegiums,
meine sehr verehrten Damen und Herren

Chers membres du Collegium,
Mesdames et Messieurs

Dear members of the Collegium,
Ladies and Gentlemen

Mit grosser Dankbarkeit für die Wahl Berns
als Tagungsort für das Collegium O.R.L.A.S.
1972 eröffne ich heute die Sitzung. Es freut
mich die grosse Zahl von Mitgliedern aus 27
Nationen, die seit Jahren bekannten Freunde
und die junge Generation begrüssen zu dürfen

ganz besonders herzlich heisse ich die beglei-
tenden Damen und Gäste willkommen. Wir
haben versucht, ein gebaltvolles Programm
aufzustellen. Besonderer Dank gilt dem Direk-
tor des Inselspitals, Herrn Dr. François Kohler
und seinem Stab für die grosszügige Gast-
freundschaft und die tatkräftige Unterstützung.
Über die besondere Stellung des seit über
600 Jahren bestehenden Inselspitals werden
Sie von ihm direkt hören.

C'est avec un sentiment profond de gratitude
vis-à-vis du Collegium d'avoir choisi Berne
comme siège pour 1972 que j'ouvre la séance.

Je suis ravi de saluer un grand nombre de membres de 27 nations, les amis connus de puis longtemps et la jeune génération. Je salue tout spécialement les dames et les hôtes. Nous avons essayé de vous offrir un programme intéressant. Je remercie tout spécialement le directeur de l'Hôpital de l'île et son équipe pour l'hospitalité généreuse et son soutien efficace. Il vous donnera tout à l'heure un aperçu sur cet hôpital, fondé il y a plus de 600 ans.

I express gratitude for the choice of Berne as the Meeting place of the Collegium O.R.L.A.S. 1972 and declare the session open.

It gives me a great pleasure to welcome the large number of members, from 27 nations, the many well-known faces and the younger generation and I especially welcome the accompanying ladies and guests. We have tried to organize an interesting program and are especially grateful to the Director of the Inselhospital and his staff for their generous hospitality and active support. He will tell you personally about the position and activities of the 600-year-old Insel Hospital.

Seit der Gründung des Collegiums 1926 ist es das 3. Mal, dass das Collegium in der Schweiz tagt. 1927 bei Fr. Nager, 1952 bei L. Rüedi in Zürich und jetzt, 1972, in Bern.

Bern gegründet 1191 von Herzog Berchtold V von Zähringen hat eine ruhmvolle Geschichte und erlangte europäische Bedeutung in der Konfrontation 1476/77 zu Karl dem Kühnen, Herzog von Burgund. Die Damen werden im Historischen Museum die Schätze der Burgunder Beute bewundern können.

Die auf der Halbinsel der Aare gebaute Stadt hat ihren baulichen Charakter bewahrt. Die Wahrzeichen der Stadt, das Münster wurde ab 1421 über fast 100 Jahre Bauzeit und das Rathaus 1406 gebaut. In den breiten Gassen entstanden die prachtvollen Brunnen während die heutigen Häuser zum grossen Teil aus der Barockzeit stammen. 1848 wurde Bern die

Hauptstadt der Schweiz. Die Universität entstand 1834 hatte aber schon ihre Vorläufer. Ich erwähne die grosse geistige Ausstrahlung des Naturwissenschaftlers und Dichters Albrecht v. Haller im 18. Jahrhundert.

Depuis la fondation du Collegium en 1926 se trouve pour la 3. fois en Suisse, 1927 chez Fr. Nager à Zurich, 1952 chez L. Rüedi à Zurich et maintenant — 20 ans plus tard — à Berne.

Berne fondée en 1191 par le duc Berchtold V de Zähringen a une histoire glorieuse et connu une période d'importance européenne en 1476/77 lors de la confrontation avec Charles le Téméraire duc de Bourgogne. Les dames visiteront les trésors du butin de Bourgogne au Musée historique.

La ville, bâtie sur la presqu'île de l'Aare, conservé son caractère médiéval. Les édifices significatifs, la cathédrale et la mairie furent commencés au début du 15^e siècle. Dans les rues larges dès le début on construisit les fameuses fontaines, alors que les maisons actuelles avec les arcades sont du baroque. En 1848 Berne fut élue comme capitale de la Confédération.

La fondation de l'université date de 1834. Il avait auparavant des écoles académiques. J cite le rayonnement spirituel du grand scientifique et poète A. von Haller au 18^e siècle.

Since the foundation of the Collegium in 1926 this is the third time that the Congress is being held in Switzerland. In 1927 with Professor Nager in Zurich, in 1952 with Professor Rüedi in Zurich and in 1972 in Berne.

Berne was founded in 1191 by the Duke Berchtold of Zähringen, has a famous history and achieved European importance during the confrontation 1476/77 with Charles the Bold Duke of Burgundy. The ladies will be able to admire the treasures of the Burgundian war in the Museum of History.

The town of Berne, built on an arm of the River Aare, has kept its architectural character. The Cathedral, started in 1471 and con-

pleted nearly 100 years later and the town hall, dating from 1406 are the landmarks of the city. Beautiful fountains were set up in the wide streets and the majority of the houses date from the baroque period. In 1848 Berne became the capital of Switzerland.

The University was established in 1834 but it had already its precursors. I am referring to the great intellectual emanation of the scientist and poet Albrecht von Haller in the 18th century.

Die Oto-Laryngologie ist in Bern in der 5. Generation. Der erste Lehrstuhlinhaber war Prof. Valentin, der Sohn des bekannten Physiologen Valentin. Damit erscheint auch der Name Alfonso Corti verbunden mit Bern. Corti verliess 1848 bei der Revolution Hyrtl in Wien kam nach Bern, arbeitete hier histologisch bei Vater Valentin, bis er dann 1850 zu Koelliker in Würzburg übersiedelte, wo seine grundlegende Arbeit über das Innenohr entstand. F. Lüscher Vater E. Lüscher Sohn und mein Lehrer L. Rüedi waren die weiteren Fachvertreter.

Heute das Collegium präkaderen zu dürfen ist für mich eine grosse Ehre und Verpflichtung, möge die Tradition der wissenschaftlichen Arbeit auf dem Boden der *amicitia sacra* blühen und Früchte tragen.

Oto-laryngologie de Berne en est actuellement à sa 5^e génération. Le premier titulaire fut le Prof. Valentin, le fils du fameux physio-

logue Valentin. Avec celui-ci le nom d'Alfonso Corti est lié à Berne. Lors de la révolution 1848 Corti a quitté Hyrtl à Vienne pour s'installer chez Valentin à Berne, où il publia plusieurs travaux histologiques. C'est de Berne qu'il se rendit en 1850 chez Koelliker à Würzburg, pour s'initier à la recherche de la structure de l'oreille interne. Le père F. Lüscher le fils E. Lüscher et mon maître L. Rüedi furent les titulaires suivants.

Présider le Collegium aujourd'hui est pour moi une honneur et une obligation. C'est mon vœu que la tradition scientifique sur la base de l'*amicitia sacra* fleurisse et continue de porter des fruits.

The ENT is in its 5th generation in Berne. The first professor was Prof. Valentin, the son of the well-known physiologist Valentin. At the same time the name of A. Corti was also connected with Berne. Corti left Hyrtl in Vienna in 1848 during the Revolution came to Berne, worked here on histology with Valentin senior until he moved to Prof. Koelliker in Würzburg in 1850 where his basic studies on the inner ear were published. F. Lüscher senior who built the clinic in 1909 E. Lüscher his son, and my teacher L. Rüedi were the next chairmen.

Today the presidency of this Collegium is for me a great honour and responsibility. I sincerely hope the tradition of scientific work and deep-rooted friendship will continue to blossom and bear fruits.

SENSITIZATION AND LOCAL APPLICATION OF ANTIBIOTICS
ON THE RESPIRATORY MUCOUS MEMBRANE

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Abstract. Local application of antibiotics to the respiratory mucous membrane in the form of drops and aerosol is more and more often used. Here the question arises of sensitization of the mucosa to the penicillin, being the strongest allergenic remedy. Experimentally we tried with aerosol to provoke the sensitization in rabbits and we observed patients who had received aerosol several times. Laboratory examinations were made in the form of basophil degranulation determinations and compared with the control group. The results are discussed.

During the last 10 years more than 60 000 aerosol inhalations with antibiotics, vasoconstrictors and cortisone have been made in our department. Everyday use of antibiotics in the form of drops applied to the nasal mucosa and the external and middle ear is well known.

As the nasal mucosa is an area of excellent permeability, the question arises as to the possibility of sensitization of the organism by the aerosol treatment. Clinical experiences have not been able to confirm this supposition which is why we have made an experimental investigation in rabbits and clinical and laboratory examinations in patients after repeated aerosol application of antibiotics. Penicillin was chosen as the most common antibiotic and the one which most frequently causes allergic reactions. Allergic reactions appear in 1 to 15 out of every 1 000 patients treated with penicillin. These reactions can be manifested as erythemo-urticarial reactions all the way to anaphylactoid shock, blood dyscrasias and even

death (the fatal reaction is about 1 in 100 000 to 150 000 treatments).

We have been interested, in the first place in the problem whether with frequent local applications of antibiotic, there is a tendency to sensitization of the organism. For our examinations crystal penicillin was chosen which only in this form is used in aerosol.

MATERIAL AND METHODS

We have divided our work into two parts. The experimental part on rabbits, and the clinical laboratory examination of the patients who received a series of 10 to 15 inhalations. Before this investigation we wanted to ascertain what quantities of penicillin exist in the blood serum with various applications.

	Minutes after application		
	15	60	180
1. 800 000 IU procaine penicillin intramuscularly	4.050	8.640	—
2. 100 000 IU benzyl penicillin intramuscularly	1.96	3.420	8.17
3. 100 000 IU with a tampon on the nasal mucosa	0.3	0.066	0.06
4. 100 000 IU of penicillin into the maxillary sinuses	0.12	0.290	0.07
5. Inhalation of 100 000 IU of benzyl penicillin	0.03	0.066	0.06

Experiments in animals

In the experimental part we used 3 groups of rabbits. In the first group we used 4 rabbits and gave them daily for 10 days, 200 000 IU

This work was supported by grant from the Council of Scientific Work of Croatia.

Table I Results of indirect test of degranulation of basophil granulocytes (ITDBG) and the test of blastic transformation of lymphocytes (TTL) in the culture before and after inhalation of penicillin

Sex	1st day								14th day								21st day							
	ITDBG				TTL				ITDBG				TTL				ITDBG				TTL			
	P		C		P		C		P		C		P		C		P		C		P		C	
	n	p	n	p	n	p	n	p	n	p	n	p	n	p	n	p	n	p	n	p	n	p	n	p
M	17	0	28	0	17	0	28	0	17	0	28	0	17	0	28	0	17	0	28	0	17	0	28	0
F	33	0	12	0	33	0	12	0	13	0	12	0	13	0	12	0	13	0	12	0	13	0	12	0
Total	50	0	40	0	50	0	40	0	30	0	40	0	30	0	40	0	30	0	40	0	30	0	40	0

P patients, C, control M, males; F females, n, normal p, pathological.

of aerosol penicillin. After 14 days the allergological tests for penicillin remained negative (intradermal, provocative with aerosol, and serological hemagglutinations according to Boyd). In the second group we used 18 rabbits which were given, preliminarily intramuscular crystal penicillin+procaine penicillin with complete Freund adjuvant (2 c.c.) After 3 weeks the tests for penicillin remained negative, including the aerosol. In the third group we infected 4 rabbits with the virus of equine influenza intranasally and intravenously. After that, for 10 days, we applied the penicillin aerosol.

The agglutination test in rabbit serum for the equine influenza virus was, after 14 days, positive in the ratio of 1 : 64. The allergological tests for the penicillin remained negative in all 4 rabbits. Thus we were not able, with 26 rabbits, to provoke an allergic reaction by applying aerosol penicillin, not even with the preliminary intramuscular application and by addition of Freund's adjuvant and the provoked virus infection.

Investigations in patients

We performed the laboratory-clinical investigation with 50 patients, who received penicillin inhalations, and as a control group we took 20 patients without inhalations or with inhalations with a physiological solution only.

In all investigated cases, including the controls, the absolute number of basophil and

eosinophil granulocytes in the peripheral blood have been determined, and ITDBG (indirect test for degranulation of basophil granulocytes) and TTL (test for lymphocyte transformation) were performed before inhalation of the antibiotic had begun. Absolute numbers of eosinophil and basophil granulocytes were determined also after 7 and 14 days, and ITDBG and TTL 14 and 21 days after inhalation. All these examinations and tests were negative in all cases (Tables I, II).

During this time clinical symptomatology was also negative. In not a single person were clinical symptoms noticed which might point to the possibility of an allergic reaction to penicillin.

There was no difference in the number of eosinophils before or after inhalation (Tables III-IV). In the repeated series of inhalations, after 1 to 11 months, in 100 cases there were no reactions (Table V).

The number of basophil granulocytes was determined according to Moore & James (1953) and that of eosinophil granulocytes by the method of Wintrobe (1961). As normal values we took for basophil granulocytes 15 to 50 cells per cubic millimetre and for eosinophil granulocytes 150 to 300 cells per cubic millimetre. We performed ITDBG according to Shelley with certain slight modifications which do not influence the result of the test (Softić & Brnobić, 1966). The test was found negative when the percentage of the damaged

Table II Absolute number of basophil and eosinophil granulocytes in the peripheral blood before and after inhalation of penicillin

Sex	1st day								14th day								21st day							
	Basophils				Eosinophils				Basophils				Eosinophils				Basophils				Eosinophils			
	P		C		P		C		P		C		P		C		P		C		P		C	
	n	p	n	p	n	p	n	p	n	p	n	p	n	p	n	p	n	p	n	p	n	p	n	p
M	17	0	28	0	17	0	28	0	17	0	28	0	17	0	28	0	17	0	28	0	17	0	28	0
F	33	0	12	0	33	0	12	0	13	0	12	0	13	0	1	0	13	0	1	0	13	0	13	0
Total	50	0	40	0	50	0	40	0	30	0	40	0	30	0	40	0	30	0	40	0	30	0	40	0

P patients C control M males, F females; n, normal p, pathological.

cells was less than 25% indefinite when the percentage of the damaged cells was between 25 and 30% and positive when the percentage of the changed basophil granulocytes was 30% or more. We performed TTL by the method of Coulson & Chalmers (1964) slightly modified by Chieragato & Faldarini (1966). In all cases we used the double control, the culture without the addition of antigens and the culture to which we added phytohemagglutinin (Bacto-phytohemagglutinin II Difco Laboratories, Detroit). The culture was estimated positive when in a complete culture (the culture with antigen) stimulation of lymphocytes increased 3 or more in the sense of blastic formation, and in the control culture (the one to which no antigen was added) there was no stimulation and when PHA was added there was a pronounced blastic transformation of lymphocytes (40 or more).

DISCUSSION

Penicillin has been studied more extensively than any other antibiotic, but the manner in which it sensitises is still the matter of lively debate and the semisynthetic penicillins have added a new dimension of antigenicity to the antigenic potential of the older compounds. In the centre penicillin is transformed into penicillemic acid which combines with tissue and yields the antigen penicilloyl protein.

This is perhaps the most significant but also the only cause for the antigenicity of penicillin.

Very small quantities of penicillin which are resorbed with aerosol give a far smaller possibility for the formation of penicilloprotein and, consequently for the possibility of the sensitization of the organism. However in this procedure relatively larger quantities of penicillin stay on the surface of the nasomucosa. According to our measurements the amount is 35 to 100 IU per cm² of the surface which facilitates the action upon the mucosa itself. We think therefore that inhalations cannot have any effect on those sections whose localization is deeper in the mucosa than a mere superficial inflammatory reaction, where there is no systematic reaction of the organism to such an infection.

Our second conclusion is that by applying aerosol we do not cause a greater likelihood of the sensitization of the organism to a definite antibiotic than the innate liability of the organism to such an allergic reaction.

Table III Eosinophiles in nasal smears before and after inhalation of penicillin

	Negative	Positive	Not taken	Total
Before inhalation	39	3	6	48
After inhalation	38	2	10	50

Table IV *Eosinophils in nasal smears from Control group during 14 day period*

	Negative	Positive	Not taken	Total
1st day	20	1	0	21
14th day	16	1	4	21

RÉSUMÉ

L'application locale des antibiotiques sur la membrane muqueuse respiratoire en forme de gouttes et aérosol s'emploie de plus en plus. Il se pose la question de la sensibilisation de la membrane muqueuse le plus souvent à la pénicilline comme le moyen allergique le plus fort. Expérimentalement, nous avons essayé de provoquer la sensibilisation à l'aérosol sur les lapins, nous avons observé les patients qui recevaient l'aérosol plusieurs fois. En même temps nous avons fait les examens en laboratoire dans forme de dégranulation basophile et nous avons fait la comparaison avec le groupe contrôle. Les résultats sont présentés.

ZUSAMMENFASSUNG

Die lokale Applikation von Antibiotika auf die Respirationschleimhaut in Form von Tropfen und Aerosol wird immer mehr angewendet. Dabei stellt sich die Frage der Sensibilisation der Schleimhaut besonders auf Penicillin, den stärksten allergenen Mittel, dar. Wir versuchten auf den Kaninchen die Sensibilisation experimentell auszuführen und beobachteten auch die Patienten bei denen der Aerosol mehrfach verwendet wurde. Dabei waren die laboratorischen Untersuchungen in Form der basophylen Degranulation durchgeführt und mit denen der Kontrollgruppe in Vergleich gestellt. Das Resultats werden aufgeführt.

REFERENCES

- Chierrigato, G. C. & Faldarini, G. 1966. La cultura *in vitro* di linfociti di soggetti con eczema professionale. *Atti Soc. Derm.* 41 115
 Coulton, A. H. & Chalmers, H. G. 1964. Separation of viable lymphocytes from human blood. *Lancet* i 468

- D- Weck, A. L. 1971. Immunochemical mechanisms of hypersensitivity to antibiotics. Solution to the penicillin allergy problem? New concepts in allergy and clinical immunology. *Excerpta Med.* pp. 208-215. Amsterdam.
 Moore, J. E. & James, H. W. 1953. A simple direct method for absolute basophil leucocyte count. *Proc. Soc. Exp. Biol. Med.* 82 601
 Sotić, M. & Brnčić, A. 1966. Indirektni test degranulacije bazofilnih granulocita. *Acta Med. Jugosl.* 20 1
 Winthrope, M. M. 1961. *Clinical Immunology* pp. 234-40. Lea & Febiger, Philadelphia.

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DISCUSSION

H. H. Neumann: Die Fragestellung von Herrn Krajinca ist vom Prinzip her sehr aktuell, denn es geht ja darum, ob wir mit einer intranasalen Medikation Arzneimittelallergie induzieren können oder nicht. Nur glaube ich, dass die gewählte Testsubstanz zur Beantwortung dieser Frage kaum geeignet ist. Die Penicilline sind niedermolekular und als echte wässrige Lösung geben sie praktisch mit der Geschwindigkeit einer intravenösen Injektion von der Schleimhautoberfläche der Nase in die Blutbahn über. Mein Mitarbeiter Dr. Midžetić konnte jüngst sowohl isotopentechnisch als auch autoradiographisch zeigen, dass bei derartigen Substanzen die Resorptionsleistung der Nasenschleimhaut sehr deutlich an die Epithelzellen gebunden ist, während überraschenderweise die Lamina propria, welche für immunologische Vorgänge in erster Linie zuständig wäre, bei niedermolekularer wässriger Lösung keine Spätersphänonome zeigt. Die Passage derartigen Flüssigkeiten geht direkt am Epithel vorwiegend interzellulär zu terminalen Strombahn, d. h. ins Blut. Damit entfällt theoretisch die Wahrscheinlichkeit, mit Penicillin in wässriger Lösung an der Nasenschleimhaut immunologische Reaktionen erzielen zu können. Deshalb meine Frage an Herrn Krajinca: Haben Sie auch andere, höhermolekulare bzw. kolloidale Medikamente verwendet oder Wirkstoffe, die eventuell ein Protein oder Protein gekoppelt waren?

Table V *Repeated inhalations: Second series (91 patients) Third series (9 patients)*

	Period (in months) between first and second, or second and third series of inhalations										
	1	1/	2	2/	3	3/	4	5	6	7	8
Patients	24	8	10	5	10	5	3	6	4	4	9
											4
											7
											1

Table II *Absolute number of basophil and eosinophil granulocytes in the peripheral blood before and after inhalation of penicillin*

Sex	1st day				14th day				21st day			
	Basophils		Eosinophils		Basophils		Eosinophils		Basophils		Eosinophils	
	P		C		P		C		P		C	
	n	p	n	p	n	p	n	p	n	p	n	p
M	17	0	18	0	17	0	28	0	17	0	28	0
F	33	0	12	0	33	0	12	0	13	0	12	0
Total	50	0	40	0	50	0	40	0	30	0	40	0

P patients C control M males, F females n, normal p, pathological.

cells was less than 25%. Indefinite when the percentage of the damaged cells was between 25 and 30% and positive when the percentage of the changed basophil granulocytes was 30% or more. We performed TTL by the method of Coulson & Chalmers (1964) slightly modified by Chiaregato & Faldarini (1966). In all cases we used the double control: the culture without the addition of antigens and the culture to which we added phytohemagglutinin (Bacto-phytohemagglutinin P, Difco Laboratories, Detroit). The culture was estimated positive when in a complete culture (the culture with antigen) stimulation of lymphocytes occurred (3 or more in the sense of blast transformation), and in the control culture (the one to which no antigen was added) there was no stimulation and when PHA was added there was a pronounced blast transformation of lymphocytes (40% or more).

DISCUSSION

Penicillin has been studied more extensively than any other antibiotic but the manner in which it sensitizes is still the matter of lively debate and the semisynthetic penicillins have added a new dimension of antigenicity to the antigenic potential of the older compounds. In the centre penicillin is transformed into penicillic acid which combines with tissue and yields the antigen, penicilloyl protein.

This is perhaps the most significant but not the only cause for the antigenicity of penicillin.

Very small quantities of penicillin which are resorbed with aerosol give a far smaller possibility for the formation of penicilloyl protein and, consequently for the possibility of the sensitization of the organism. However in this procedure relatively larger quantities of penicillin stay on the surface of the nasal mucosa. According to our measurements they amount to 35 to 100 I.U. per cm² of the surface which facilitates the action upon the mucosa itself. We think, therefore, that inhalations cannot have any effect on those infections whose localization is deeper in the mucosa than a mere superficial inflammatory reaction where there is no systematic reaction of the organism to such an infection.

Our second conclusion is that by applying aerosol, we do not cause a greater likelihood of the sensitization of the organism to a definite antibiotic than the innate liability of the organism to such an allergic reaction.

Table III *Eosinophiles in nasal smears before and after inhalation of penicillin*

	Negative	Positive	Not taken	Total
Before inhalation	39	5	6	40
After inhalation	38	2	10	40

Table IV *Eosinophils in nasal smears from Control group during 14 day period*

	Negative	Positive	Not taken	Total
1st day	20	1	0	21
14th day	16	1	4	21

RÉSUMÉ

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REFERENCES

- Chiaregato, G. C. & Faldutini, G. 1966 La cultura in vitro di *Bafocti* di soggetti con eczema professionale. *Minerva Derm* 41 115
- Coulton, A. S. & Chalmers, H. G. 1964 Separation of viable lymphocytes from human blood *Lancet* i 468.
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- Softić, N. & Brnobić, A. 1966. Indirektni test degranulacije bazofilnih granulocita. *Acta Med Jugosl* 20 1
- Whitroba, M. M. 1961 *Clinical hematology* pp. 234-40. Lea & Febiger Philadelphia.
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	Period (in months) between first and second, or second and third series of inhalations											
	/	1	1/2	2	2 1/2	3	3 1/2	4	5	6	7	8
Patients	24	8	10	5	10	5	3	6	4	4	9	4
											7	1

T. Leegard. It is my sincere conviction that in 99 out of a 100 times local application of antibiotics is useful only to influence our normal, useful mixture of micro-organisms in the nose and throat and which, besides possible sensitization, is resulting only in destroying the ecology of the micro-organisms.

H. C. Andersen. I think it is very important that Mr Krajina has shown us that local application of penicillin in the nose is not dangerous, but I think he owes us the proof that such application has any therapeutic effect.

Z. Krajina (Reply). Our intention was to prove that it is possible to sensitize the organism more effectively

through the application of aerosol penicillin. We were surprised at the small quantity of penicillin in the blood after the inhalation. Perhaps that was due to the scattering of penicillin over the respiratory mucosa after the application of the aerosol. We used in our work only antibiotics combined with vasoconstrictors and cortisone and I have not tried any substances with high molecular weight. About the value of aerosols in clinical practice: only superficial infection of the respiratory mucosa can be influenced by aerosol; otherwise it is necessary to use antibiotics systemically or in combination with the aerosol.

MORPHOLOGISCHE UNTERSUCHUNGEN ZUR RETIKULIERUNG
DES TONSILLENEPITHELIS

P. Falk und W. Mootz

Aus der Hals-Nasen-Ohrenklinik der Universität des Saarlandes, Homburg/Saar BRD

Abstract: Im Tierversuch läßt sich die Retikulation des Kryptenepithels durch Gaben von Cortison und Desoxycorticosteron variieren. Das Ausmaß der retikulierten Epithelschicht geht postnatal mit der Ausbildung der Sekundarfollikel parallel. Die Retikulierung wird als ein physiologischer Vorgang angesehen. Elektronenmikroskopisch stellt sich der Retikulierungsvorgang wie folgt dar: zunächst gelangen Lymphocyten zwischen zwei Epithelzellen durch die Basalmembran in tiefe Epithelschichten. Weitere Zellen drängen durch die so entstandenen Lücken in das Epithel ein, wo sie dann zwischen fein ausgezogenen Fortsätzen der Epithelzellen in einem Odemas liegen. In den mittleren und oberen Epithelschichten finden sich ferner Lymphoblasten in Teilung. Gelegentlich beobachtet man Lymphocyten über intakter Basalmembran. Beim Menschen wird das Retikulierungsbild von entzündlichen Veränderungen überlagert. Die morphologischen Befunde werden in Beziehung zur Tonsillenfunktion gesetzt. Auf das Vorkommen von Langerhans-Zellen im Epithel der menschlichen Gaumenmandel wird eingegangen.

Neuere Ergebnisse über die immunologische Funktion der Tonsilla palatina lassen es als gesichert erscheinen, daß dieses lymphoepitheliale Organ eine wesentliche Rolle bei der Immunisierung des gesamten Organismus spielt (Naumann 1957 Meyer zum Gottesberge & Koburg, 1967 Koburg, 1970). Diese Erkenntnisse veranlassen uns, auch elektronenmikroskopisch das Epithel der Gaumenmandel unter besonderer Berücksichtigung seiner Retikulierung zu analysieren. Im Rahmen dieser Untersuchungen fanden wir erstmals Langerhans-Zellen im Epithel der Gaumenmandel des Menschen.

Während unsere Kenntnisse über die Ultrastruktur des retikulierten Kryptenepithels, das für die spezifische Tonsillenfunktion im Rahmen immunologischer Vorgänge von so großer

Bedeutung ist, nur spärlich sind, finden sich lichtmikroskopisch zahlreiche Untersuchungen (Stöhr 1882, 1884 Dietrich 1923 Watzka, 1932, Schwarz, 1953 Kelemen, 1954 Boenninghaus, 1955 Yamaguchi, 1960 Fioretti, 1961 Grau, 1965). Die Schwierigkeiten bei der Deutung dieses Vorganges waren vor allem darin begründet, daß fast immer operativ entfernte Tonsillen von Menschen verwendet wurden, bei denen das physiologische Retikulierungsbild durch pathologische Veränderungen bei chronischer Tonsillitis überlagert war.

Den Retikulierungsvorgang am Kryptenepithel der Tonsilla palatina versuchten wir daher an nicht entzündlich veränderten Tonsillen weiter abzuklären. Zu diesen Untersuchungen bot sich uns die Kanchentonsille an an der wir schon früher bei tierexperimentellen Studien mit Nebennierenrindenhormonen das Ausmaß der Retikulierung des Tonsillenepithels modifizieren konnten (Falk & Schätzle, 1962 Schätzle & Falk, 1962).

MATERIAL UND METHODIK

1 *Tierexperimentelle Untersuchungen zum Retikulierungsvorgang am Kryptenepithel der Tonsilla palatina nach Injektion von Cortison Methylprednisolon und Desoxycorticosteron (DOCA):*

a) *Cortison.* 6 Kaninchen (Rasse „Grau Silber“ 31 Tage alt) erhielten an 6 aufeinanderfolgenden Tagen je 40 mg/kg Körpergewicht subcutan. 2 Tage danach Dekapitation. 5 Kontrolltiere.



Abb. 1 (a) Kontrolltier. Deutliche Retik. (b) 6 \times 40 mg Cortison. Atrophie des Sekundärfollikels. (c) 4 \times 15 mg DOCA. Cortisonbedingte Atrophie des lymphatischen Gewebes aufgehoben. Kraftige Retikulierung. (d) 4 \times 15 mg DOCA. Epithel über den Strecken durchgehend rekonstruiert. Dicht nebeneinander liegende Sekundärfollikel mit Pfortappe.

b) *Methylprednisolon*. 9 Kaninchen (Rasse „Grau Silber“ 10 Monate–2 Jahre alt). 6 \times 20 mg/kg Körpergewicht. 7 Kontrolltiere. Töten der Tiere 2 Tage nach der letzten Injektion durch Einspritzen von Chloroform in die Nasenhöhle.

c) *Desorvcorticosteron*. 14 Kaninchen (Rasse „Grau-Silber“ 28–50 Tage alt) wurden 4 \times 15 mg/kg Körpergewicht subcutan injiziert. Dekapitation 2 Tage nach der letzten Injektion 7 Kontrolltiere.

d) *Cortison und Desorvcorticosteron*. 11 Kaninchen (Rasse „Deutscher Widder“ 10 Monate alt) wurden 6 \times 40 mg/kg Körpergewicht Cortison verabreicht und nach einer Pause von 2 Tagen 4 \times 15 mg/kg DOCA. 11 Kontrolltiere (unbehandelt, 6 \times 40 mg Cortison, 4 \times 15 mg DOCA). Töten der Tiere 2 Tage nach der letzten Injektion.

2. Tonsillen von 12 Kindern, die wegen hyperplastischer chronischer Tonsillitis tonsillotomiert wurden.

3 Untersuchungstechnik

a) *Lichtmikroskopie*. Fixation in 10 ligen neutralem Formalin, Paraffineinbettung. Färbungen: HE, Goldner, Aldehydthionin nach Specht (1970), Perjodsäure-Blaulit, Aldehydthionin (PBA-Reaktion) nach Specht, PBA-Methode nach Vorbehandlung mit Diastase, Toluidinblau (pH 3–4).

b) *Elektronenmikroskopie*. Immersionsfixation in 6,25% ligen eisgekühltem Glutaraldehyd in Sörensen-Phosphatpuffer (pH 7,4) über 1¹/₂ Stunden. Zweistündige Nachfixation in 1% ligen Osmiumsäure in Veronal-Acetat-Puffer (pH 7,4) mit Zusatz von 4,5% Saccharose (Cath-

Herrn Dr. Münke (Pathologisches Institut) danken wir für die Unterstützung bei den elektronenmikroskopischen Untersuchungen.

phie des lymphatischen Gewebes. Sporadisch rekonstruiertes kryptonepithel (c) 6 \times 40 mg Cortison und 4 \times 15 mg DOCA. Cortisonbedingte Atrophie des lymphatischen Gewebes aufgehoben. Kraftige Retikulierung. (d) 4 \times 15 mg DOCA. Epithel über den Strecken durchgehend rekonstruiert. Dicht nebeneinander liegende Sekundärfollikel mit Pfortappe.

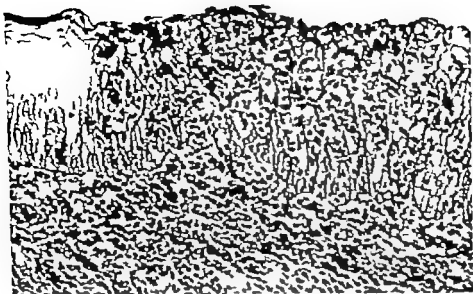


Abb 2 Erwachsener Kontrolltier PBA-Färbung nach Specht. Basalmembran unter dem retikulierten Epithel unterbrochen. 260

field, 1957) Nach Aralditeinbettung wurden die Ultradünnschnitte (Ultramikrotom Om U 2, Fa. Reichert) mit Uranylacetat (Watson, 1958) und Bleicitrat (Reynolds, 1963) kontrastiert. Auswertung mit dem Zeiss EM 9 S2. Kontrollschnittfärbungen. Methylenblau (Unna), Movat Versilberung.

4 Zum Nachweis der Langerhans-Zellen im Epithel der menschlichen Gaumenmandel kamen 22 Tonsillenpaare, die wegen chronischer Tonsillitis ausgeschält wurden, zur Untersuchung.

a) *Lichtmikroskopische Untersuchungen.* HE und Melaninfärbung nach Fixation des Gewebes in neutralem 10% igem Formalin. Osmium-Zinkjodid-Methode nach Malliet (1959). Semidünnschnittfärbung mit Methylenblau (Unna).

b) *Elektronenmikroskopie* Fixation der auf geschnittenen Tonsillen über 12 Stunden in 6,25% igem eisgekühltem Glutaraldehyd in Sörensen-Phosphatpuffer (pH 7.4) Gezielte Ent-

nahme von Oberflächen und Kryptenepithel. Weiterverarbeitung wie unter 3b beschrieben.

ERGEBNISSE

Lichtmikroskopische Befunde an der Kaninchentonsille

Kontrolltiere

Die Tonsilla palatina besitzt meist nur eine zentral gelegene Krypte. Bei den jungen Tieren sind die extrafollikulär gelegenen Lymphocyten mäßig dicht angeordnet. Die Sekundärfollikel liegen nahe beieinander. Die Retikulation des Epithels ist stärker über den Sekundärfollikeln ausgeprägt, geht aber auch auf die interfollikulären Areale des Epithels über.

Bei den erwachsenen Kontrolltieren sind die zahlreichen Sekundärfollikel mit Lymphocytenkappen und hellen Zentren ebenso gut ausgebildet wie das extrafollikuläre lymphatische Gewebe. Im Gegensatz zu den Jungtieren ist das Kryptenepithel höher. Die Retikulation ist nur auf die Zone über den Kopf

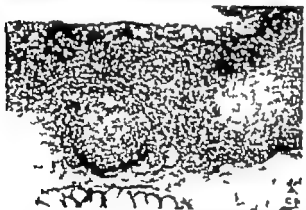
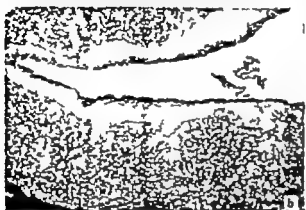


Abb. 1 (a) Kontrolltier: Deutliche Retikulierung über den Sekundärfollikeln. (b) 6 x 40 mg Cortison: Atro-

b) *Methylprednisolon* 9 Kaninchen (Rasse „Grau Silber“ 10 Monate – 2 Jahre alt). 6 20 mg/kg Körpergewicht, 7 Kontrolltiere. Töten der Tiere 2 Tage nach der letzten Injektion durch Einspritzen von Chloroform in die Nasenhöhle.

c) *Desoxycorticosteron*. 14 Kaninchen (Rasse „Grau Silber“ 28–50 Tage alt) wurden 4 x 15 mg/kg Körpergewicht subcutan injiziert. Dekapitation 2 Tage nach der letzten Injektion 7 Kontrolltiere.

d) *Cortison und Desoxycorticosteron*. 12 Kaninchen (Rasse „Deutscher Widder“ 10 Monate alt) wurden 8 x 40 mg/kg Körpergewicht Cortison verabreicht und nach einer Pause von 2 Tagen 4 x 15 mg/kg DOCA. 10 Kontrolltiere (unbehandelt 6 x 40 mg Cortison, 4 x 15 mg DOCA) Töten der Tiere 2 Tage nach der letzten Injektion.

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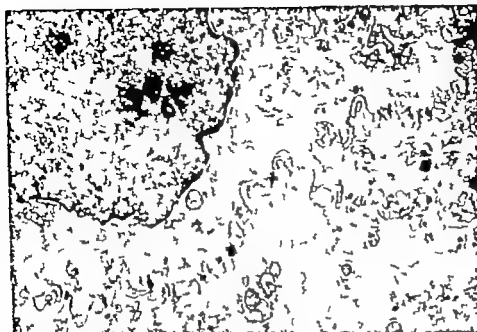


Abb. 6. Intermediärschicht. Kern abgerundet. Tonofibrillen im Cytoplasma. 10 000.

Das retikulierte Kryptenepithel

Über den Sekundärfollikeln treten mehr und mehr Lymphocyten, Plasmazellen und Makrophagen unter der Basalmembran auf und drängen hier die Kollagenfasern auseinander. Einzelne Zellen stülpen die Basalmembran ein, so daß Pseudopapillen entstehen die licht mikroskopisch dem Bild der gewellten Basalmembran entsprechen (Abb. 8). Demgegenüber sind die echten Papillen gut abgrenzbar da sie eine zentrale Kapillare enthalten, deren Endothelzellen an der Außenseite eine eigene Basalmembran besitzen und von einem schmalen Stroma umgeben sind (Abb. 9). Ist das Gefäß im Epithel quergetroffen dann erkennt man die echte Papille an dem schmalen Bindegewebsmantel mit Kollagenfasern die das Gefäß umgeben. Kapillaren ohne Bindegewebsmantel kommen im Epithel nicht vor.

Die Basalmembran ist an vielen Stellen in unterschiedlicher Breite unterbrochen (Abb. 10). Durch diese Lücken gelangen Rundzellen zwischen die basalen Epithellen, die auseinandergedrängt werden. Die Epithellen werden

schmäler und stark ausgezogen so daß ein weites Maschenwerk vorliegt (Abb. 11). Zwischen diesen schmal ausgezogenen zusammenhängenden Epithelfortsätzen liegen die eingeschleusten Zellen in einem Ödemsee. Bei den Zellen handelt es sich um Lymphocyten, Plasmazellen und Makrophagen. In den beiden oberen Zellschichten liegen auch Lymphoblasten, von denen sich einzelne in Teilung befinden. Plasmazellen sind selten vorhanden. Hat die Retikullierung alle Epithelschichten erfaßt, so ist die oberste Epithelzellschicht schmal ausgezogen und vorgebuckelt (Abb. 12).

Das Retikullierungsbild der kindlichen Gaumenmandel

Lichtmikroskopisch lag immer eine unspezifische chronische Kryptentonsillitis vor. Die Retikullierung war über den Sekundärfollikeln am stärksten ausgeprägt.

Elektronenmikroskopisch entsprechen der Epithelaufbau und das Retikullierungsbild den Befunden beim Kaninchen. Quantitativ ist der Umfang der Retikullierung jedoch bedeutend



Abb 7 Stratum superficiale. Zellen abgeflacht. Verzahnung der Zellen durch kurze Fortsätze, die auch zum Kryptalumen hin vorhanden sind. $\times 4000$.

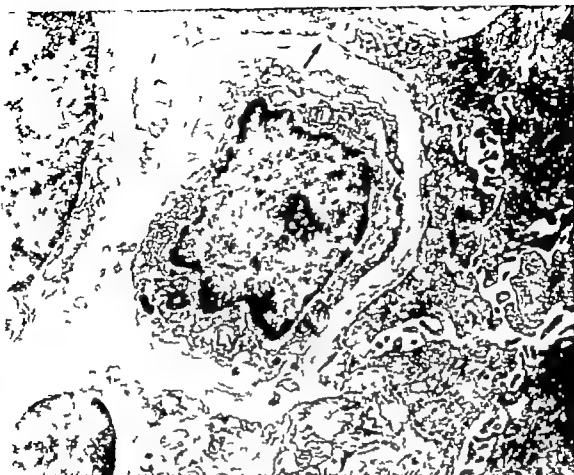
stärker. Dabei ist die Basalmembran oft nur noch bruchstückhaft vorhanden. Wenige ausgezogene Epithelzellen liegen zwischen massenhaften Rundzellen. Vermehrt kommen Plasmazellen vor; sie umsäumen häufig breit die apillaren innerhalb der Papillen (Abb 13). Jeht selten finden sich neutrophile Granulozyten im retikulierten Epithel. Auch Lymphoblasten in Teilung kommen vor.

Elektronenmikroskopischer Nachweis von Langerhans-Zellen im Oberflächen- und Kryptenepithel der menschlichen Gaumenmandel

Im Osmium-Zinkjodid-Präparat sieht man im Oberflächen- und Kryptenepithel suprabasal gelegene tiefschwarze dendritenartig verzweigte Zellen, die dicht beieinanderliegen und mit ihren Ausläufern zu anastomosieren scheinen (Abb 14). Sie können mit den Langerhans-Zellen der Epidermis verglichen werden. Nur wenige dieser Zellen liegen im Stratum basale, aber keine im darunter liegenden subepithelia-

len Gewebe. Die Melaninfärbung ist negativ. Im methylenblaugefärbten Kontrollschnitt haben diese Zellen ein helles Cytoplasma und einen stark eingebuchteten Kern.

Elektronenmikroskopisch besitzen diese Zellen ein gering elektronendichtes Cytoplasma (Abb 15). Sie sind nicht durch Desmosomen mit den benachbarten Zellen verbunden. Der Kern hat tiefe Einbuchtungen, ein lockeres Chromatingerüst und einen deutlichen Nukleolus. Im hellen Cytoplasma finden sich stäbchenförmige Mitochondrien vom Crista Typ. Der Golgi-Apparat und das endoplasmatische Retikulum sind unauffällig. Lysosomen und Zentrionen kommen ebenfalls vor. Zwischen Golgi-Feld und der Zellmembran sowie meist nur auf eine Zellhälfte beschränkt liegen stäbchenförmige, seltener auch tennischlägerartige Strukturen und rasterartige Areale (Abb. 16). Die stäbchenförmigen Langerhans-Zell-Granulatsitzen eine deutliche Querstreifung mit einer Periodik von 90 Å. Im Krypten- und Oberflächenepithel wurden die Langerhans-Zellen erstmals in Teilung gesehen. Seltener



8 Pseudopapille durch Vorbockung der intraepithelialen Basalmembran (→). Lymphocyt (Ly). 11 750.

nach in den peripheren Zellausläufern kom-
pakt, röhrenförmig gekrümmte Stäbchen dicht neben-
einander

DISKUSSION

Die Retikulierung des Epithels wird heute
als eine spezifische, physiologisch wesent-
liche Eigenschaft des Tonsillengewebes und
nicht als eine Antwort auf entzündliche Reize
angesehen (Stöhr 1884 Berggren & Hellman,
1930 Schwarz, 1953 Moser & Braun, 1954
Koretz, 1961 Falk, 1963 Koburg, 1970
Mann, 1970). Dafür spricht auch, daß die
Tonsillenmandeln schon beim 6 Monate alten
Meerschweinchen retikuliertes Epithel enthalten. Auch
bei unseren jungen Kaninchen waren niemals

Zeichen der Entzündung, aber immer reti-
kulierte Epithelabschnitte vorhanden.

Wie schon bekannt, unterscheidet man am
nicht retikulierten Epithel elektronenmikro-
skopisch eine Basalzellschicht, eine Interme-
dialschicht und eine oberflächliche Schicht
(Kato 1964 Nosaka, 1966) Diese Autoren
fanden zur Basalzellschicht hin zunehmend
weiter werdende Interzellularräume. In optimal
fixierten Gewebsschnitten bleibt jedoch die
Erweiterung des interzellulären Spaltraumes
aus. Da man auch oft perinukleäre Spalträume
in den Zellen beobachtet, zwischen denen weite
interzelluläre Spalträume liegen, besteht der
dringende Verdacht, daß es sich um Schrump-
fungsartefakte handelt. Man kann auch nicht



Abb. 9. Echte Bindegewebspapille mit Kapillare (C). Basalmembran (---) nicht unterbrochen. 5000

Nosaka zupflachten der annimmt daß die in die basal gelegenen Interzellularräume hineinragenden Zellfortsätze eine Resorptionsfunktion besitzen denn es fehlen Resorptionsvaku-

olen und ein gut ausgebildeter lysosomaler Apparat.

Da bisher nur lichtmikroskopische Befunde vom retikulierten Epithel vorlagen, waren



5b 10 Retikulation. Basalmembran unterbrochen
 →). Lymphocyten dringen in das Epithel ein. 4 250

Aussagen über das Verhalten der elektronen-
 mikroskopisch sichtbaren, für den Retikule-
 rungsvorgang wichtigen Basalmembran kaum
 möglich. Die bisher angewandten Bindegewebs-
 färbungen stellten neben den Kollagenfasern
 auch ungeformte Bindegewebsstrukturen dar.
 (Der Aldehydhionin-Methode nach Specht
 1970) wird der Verlauf der Basalmembran
 deutlicher dargestellt, da die Kollagenfasern
 nicht angefärbt werden. Mit dieser Methode
 läßt sich die Lage der unmittelbar subepithelial

gelegenen oder in den Bindegewebspapillen
 zum Epithel hin verlaufenden Kapillaren gut
 beurteilen. Die vieldiskutierte Frage, ob in
 nicht retikuliertem Epithel Kapillaren vorkom-
 men (Naumann, 1954) oder nicht (Boenning-
 haus, 1955) und wie diese sich bei der Reti-
 kulierung des Epithels verhalten, konnte je-
 doch nur elektronenmikroskopisch geklärt
 werden.

Ultrastrukturell kommen nach unseren Be-
 funden keine Kapillaren im nicht retikulierten



Abb. 11 Lymphocyten (Ly) in einem Odemsee zwischen schmal ausgezogenen Fortsätzen der Epithelzellen (E). 4500

Gewebe bei erhaltener Basalmembran vor. Die in quer und längs getroffenen Papillen liegenden Kapillaren haben eine eigene unter den Endothelzellen verlaufende Basalmembran und sind von einem Bindegewebsmantel mit Kollagenfasern umgeben. Erst daran anschließend liegt die Basalmembran der basalen Epithelzellen. Die Kapillaren sind somit dem extra epithelialen Gewebe zuzuordnen.

Die Kapillaren im retikulierten Epithel grenzen ebenfalls niemals direkt an die Epithelzellen, wie dies von Drabe & Girschik (1963) behauptet wird: sie werden immer von Bindegewebsfasern umgeben. Die Basalmembran ist nur zwischen den Epithelzellen unterbrochen, d. h. daß auch im retikulierten Epithel die

Basalzellen stets durch eine Basalmembran gegenüber dem Bindegewebsmantel der Kapillaren abgegrenzt werden. Auch der Behauptung von Schwarz (1953), daß am Beginn der Retikulierung von der Papille aus eine Entsprössung von Kapillaren durch die Basalmembran erfolgen könne, wir nicht zustimmen, da wir niemals dort Kapillarsprossungen beobachten konnten.

Am Epithel beobachteten wir bei der Retikulierung elektronenmikroskopisch veränderte Zustandsbilder: wellenförmig verlaufende Basalmembran, Vorhandensein von Bruchstücken oder streckenweises Fehlen der Basalmembran, manchmal Intaktheit der Basalmembran trotz ausgeprägter Retikulierung.



Abb. 12 Lymphocyten (Ly) und Lymphoblasten (Lb) im Stratum superficiale. 5 000.

Wir schließen aus dem gleichzeitigen Neben- einander derartiger Bilder auf zeitlich hinter einandergeschaltete, unterschiedlich weit fort geschrittene Retikulierungsstadien.

Die Retikulierung beginnt mit der Wellung der Basalmembran. Meist liegen unter der eingebuchteten Basalmembran Lymphocyten (Pseudopapillen). Dann bricht zwischen den basalen Epithelzellen die Basalmembran auf. Durch die so entstandenen Lücken dringen Lymphocyten in das Epithel ein. Durch das Auseinandergehen der Epithelzellen entstehen breitere Lücken. Lymphocyten und Plasma zellen sowie deren Vorstufen liegen dann in kleinen Gruppen in einem Odemsee, so daß die Epithelzellen weit auseinandergeschoben, nur durch lang auseinandergezogene Zellfortsätze netzartig miteinander verbunden sind.

Der Odemsee entsteht nach unserer Meinung durch Flüssigkeitsverschiebung aus dem lymphatischen Parenchym in das Epithel bei unterbrochener Basalmembran. Das epitheliale Maschenwerk erscheint geeignet, die Lymphocyten im Epithel festzuhalten so daß längere Kontakte mit Antigenen ermöglicht werden.

Im retikulierten Epithel fanden wir niemals zwischen den Rundzellen Bindegewebsfasern. Nach Aufbruch der Basalmembran war auch niemals ein Kriterium für das Vorwachsen der Papillen, noch eine explosionsartige Durchwanderung über den Sekundärfollikeln zu sehen, wobei sich dann lymphatische Zellen und Bindegewebsfasern in das Kryptolumen entleeren sollen, wie dies Kelemen (1943 1954) sah.

Bei den tierexperimentellen Untersuchungen



Abb 13 kindliche Tonsille Querschnittene Papille mit kapillare (C), die nur von Plasmazellen (Pc) umgeben ist. $\times 4500$

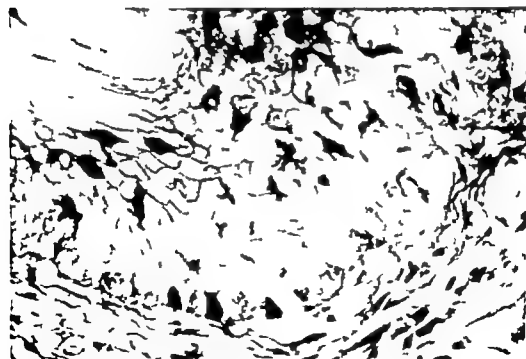


Abb 14 Menschliches Gaumenmandel-epithel Osmium-Zinkjodid Methode Suprabasal gelegene Dendriten-Zellen. $\times 40$

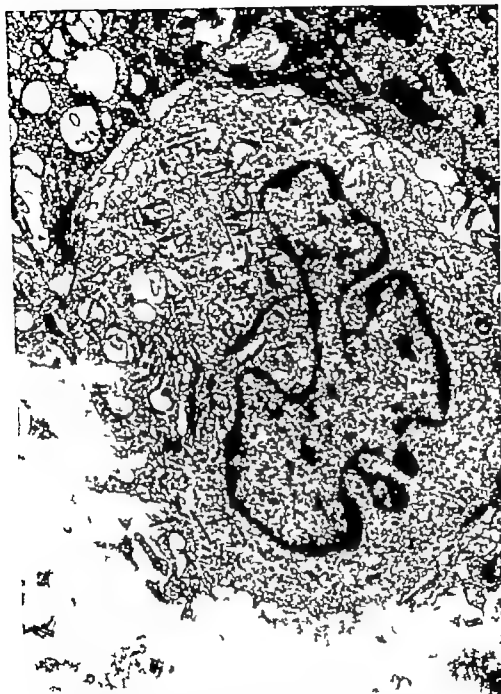


Abb. 15 Langerhans-Zelle mit gelapptem Zellkern (N) und hellem Cytoplasma. Mehrere stäbchenförmige

Strukturen (→) zwischen Golgi-Feld (G) und Zellmembran. Desmosomen fehlen. 11 000.

mit Nebennierenrindenhormonen konnten wir Ausmaß der Retikulation modifizieren, daß eine Entzündung mit im Spiel war. Wir konnten so den Retikulierungsvorgang

unverfälscht darstellen. Die Versuche mit DOCA zeigten auch, daß die Stärke der Retikulation parallel zur Ausbildung der Sekundärfollikel verläuft. Das Retikulierungsbild war

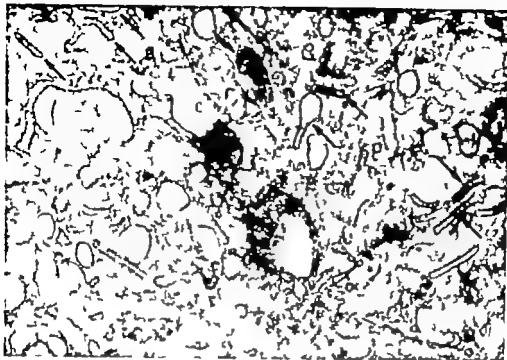


Abb. 16 Langerhans-Zell-Granula. Tennisballiger artige und silbenförmige Anschlüsse sowie rasterartige Areale (→). $\times 4.500$

aber gleich. Die Retikulierung stellt sich an den Tonsillen von Mensch und Kaninchen gleich dar. Bei den chronisch entzündeten menschlichen Tonsillen wird jedoch das Bild von entzündlichen Reaktionen überlagert.

Unsere Ergebnisse am retikulierten Epithel der Tonsille lassen sich mit den neueren Ergebnissen der immunologischen Funktion der Tonsille in Beziehung setzen. Das retikulierte Epithel ist ein wichtiges Glied in einem miteinander verbundenen Immunsystem (Keimzentrum, Lymphocytenkappe subepitheliale Zone retikuliertes Epithel). Das Plattenepithel der Tonsillenkrypte hat für das lymphatische Parenchym die Funktion, den Lymphocyten eine genügende Kontaktzeit mit Antigenen und die Möglichkeit zur Rückwanderung zu bieten.

Sowohl im krypten wie im Oberflächenepithel der Gaumenmandel konnten wir erstmals Langerhans Zellen nachweisen. Der elektronenmikroskopische Nachweis von Langerhans-Zell-Granula ist hierfür beweisend. Die vielfältige Struktur der Granulaanschnitte erklärt sich durch den dreidimensionalen Auf-

bau des Langerhans-Zell-Granulum (Wolff, 1967 b; Sagebiel & Reed, 1968; Itho, 1970).

Die Zellen sollen mesenchymaler Herkunft sein, zumal der Nachweis von Langerhans-Zell-Granula in Histocyten bei der Histocytosis X erbracht wurde (Basset et al. 1965; De Man 1968; Hashimoto & Tarnowski, 1969; Caputo & Ghanotti 1970). Außerdem enthalten die Zellen Enzyme, die typischerweise in Bindegewebszellen gefunden werden (Wolff 1967 a) und können phagozytieren (Breathnach & Wylle, 1965; Mishima, 1966). Eine Abstammung vom Nervensystem ist unwahrscheinlich, da in der Epidermis elektronenmikroskopisch niemals ein direkter Kontakt zwischen Nerven und Langerhans-Zellen beobachtet wurde (Breathnach, 1965; Zelickor 1966) und es zu keinerlei Veränderungen in den Zellen in pathologisch denervierter Haut kommt (Breathnach et al., 1962). Gegen eine melanocytaire Herkunft spricht das Fehlen von Prämelanosomen und Melanosomen im Cytoplasma der Langerhans-Zelle und daß im Tonsillenepithel des Menschen keine Melanocy-

vorkommen. Auch die Tatsache, daß in pigmentierten und unpigmentierten Hautabschnitten die Zahl der Langerhans-Zellen pro Flächeneinheit konstant bleibt, die der Melanocyten aber erheblich schwankt, untermauert dies (Wolff 1967 a)

Langerhans-Zellen konnten außer in der Epidermis u. a. auch in der Gingiva (Schroeder & Theilade, 1966) und im Cervix Epithel (Hackemann et al., 1968) nachgewiesen werden, so daß ihr Vorkommen in der Tonsille als ein für dieses Organ unspezifischer Befund angesehen werden muß. Da die mit Histiocytosis X bezeichneten Krankheitsbilder (Morbus Hand-Schüller-Christian, Morbus Letterer-Siwe und Eosinophiles Granulom) auch die Tonsillen befallen können (Brunner 1951), sollte man sich davor hüten, beim Nachweis von Langerhans-Zell-Granula im Tonsillenepithel falsche diagnostische Schlüsse zu ziehen, weil diese Granula schon unter „Normalbedingungen“ in der Tonsille vorkommen.

RÉSUMÉ

On peut faire varier la formation du réticulum de l'épithélium des cryptes en administrant de la cortisone ou de la dexaméthysone. La dimension des coupes épithéliales réticulées évolue, après la naissance, parallèlement à la formation des follicules secondaires. La formation du réticulum est considérée comme un processus physiologique. Au microscope électronique le processus de formation du réticulum se présente de la manière suivante: les lymphocytes pénètrent d'abord entre deux cellules épithéliales à travers la membrane basale dans les couches profondes épithéliales. D'autres cellules pénètrent dans l'épithélium par l'orifice ainsi formé. Chez l'homme au schéma de la formation du réticulum se superposent des transformations inflammatoires. Les données morphologiques sont en relation avec la fonction tonsillaire.

SUMMARY

In animal experiments reticulation of the crypt epithelium can be varied by administration of cortisone and dexamethasone. The amount of reticulated epithelium in the postnatal period is found to be correlated by the development of secondary follicles. Reticulation is regarded as a physiologic occurrence. By electron microscopy the pattern of reticulation appears to be as follows: lymphocytes migrate between two epithelial cells through the basement

membrane into deeper epithelial layers. Other cells advance through these gaps into the epithelium, where they are found lying between delicate processes of epithelial cells surrounded by edema fluid. In the central and superficial epithelial layers, in addition, lymphoblasts are found in mitosis. Occasionally lymphocytes are seen above an intact basement membrane. In man, inflammatory changes are superimposed on the pattern of reticulation. The morphological findings are brought into relation with tonsillar function. The occurrence of Langerhans-cells in the epithelium of human palatine tonsils is taken into consideration.

LITERATUR

- Basset, F., Nazeif, C., Mallet, R. & Turlet, J. 1965. Nouvelle mise en évidence par la microscopie électronique des particules d'allure virale dans une seconde forme clinique de l'histiocytose X, le granuloma éosinophile de Fox. *C R Acad Sci (Par.)* 261 5719.
- Berggren, S. & Hellman, T. 1930. Die chronische Tonsillitis. Ein Beitrag zur Lösung des Tonsillenproblems. *Acta Otolaryng. (Stockh.)*, Suppl. 12.
- Boemmigk, H. 1955. Über den Retikullierungsvorgang am Epithel der Tonsille und die dabei auftretenden Veränderungen an der Glitterfaserstruktur. *Z Laryng Rhinol Otol* 34 289.
- Breathnach, A. S. 1965. The cell of Langerhans. *Int Rev Cytol* 18 1.
- Breathnach, A. S., Birbeck, M. S. & Everall, J. D. 1962. Observations on Langerhans-cells in Leprosy. *Br J Derm* 74 243.
- Breathnach, A. S. & Wyllie, L. M. 1965. Melanin in Langerhans cells. *J Invest Derm* 45 401.
- Brunner, H. 1951. Eosinophilic granuloma of mouth, pharynx, and nasal passages. *Otol Surg* 4 623.
- Caputo, R. & Ghazizadeh, M. 1970. Langerhans-cells in histiocytosis X vincoblastine-induced changes. *VII Int Congr Micr Electronique Grenoble*, p. 847.
- Canfield, J. B. 1957. Effects of varying the vehicle for O_2O_3 in tissue fixation. *J Biophys Biochem Cytol* 4 877.
- De Man, J. C. H. 1968. Rod-like tubular structures in the cytoplasm of histiocytes in "Histiocytosis X". *J Path Bact* 95 123.
- Dietrich, A. 1923. Das pathologisch-anatomische Bild der chronischen Tonsillitis. *Z Hals Nasen Ohrenheilk* 4 429.
- Drabe, J. & Grischak, L. 1963. Zur Blutgefäßdarstellung in der menschlichen Gaumenmandel. *Z Laryng Rhinol Otol* 42 573.
- Falk, P. 1963. Entwicklungsgeschichte, Anatomie, Mißbildungen, Physiologie und Pathophysiologie des Rachens (einschließlich Tonsillen). In: *Handbuch der Hals-Nasen-Ohren-Heilkunde* hrsg. von J. Berendes, R. Link u. F. Zöllner Bd. II/Teil 1 Georg Thieme Verlag, Stuttgart.
- Falk, P. & Schatzle, W. 1962. Das histologische Bild der Tonsillen des erwachsenen Kaninchens nach

- Injektionen von Desoxycorticosteron. *Arch Ohr Nas Kehlkopfheilk* 179 431
- Florenti, A. 1961 *Die Gaumenmandel*. Darstellung der Biologie und Pathopathologie. Georg Thieme Verlag, Stuttgart.
- Gran, H. 1965 Lymphocyte and Mandible epithel. *Anat Anz* 116 370
- Hackemann, M., Grubb, C. & Hill, K. R. 1968. The ultrastructure of normal squamous epithelium of the human cervix uteri. *J Ultrastruct Res* 23 443
- Hoshimoto, K. & Tarnowski, W. M. 1968 Some new aspects of the Langerhans-cell. *Arch Derm* 97 450
- Ito, M. 1970 Fine structure of Langerhans-cell granule in pathologic histocytes as observed by specimen tilt electron microscopy. *11 Int Congr Micro Electronica Grenoble*, p. 443
- Kato, S. 1964 a. Electron microscopic studies of the epithelium of shallow part of hypertrophic intra-torcular cleft in man. *Tokushima J Exp Med* 11 177
- 1964 b. Electron microscopic studies of the epithelium of shallow part of the intratorcular cleft in chronic tonsillitis in man. *Tokushima J Exp Med* 11 134
- Kellemen, H. 1943 Pathway of the tonsillar lymphocyte. *Arch Otolaryng (Chic.)* 38 433
- 1954 Histological test of tonsillar activity based on the reticuloepithelial interplay. *Arch Otolaryng (Chic.)* 59 263
- Koburg, E. 1970 Die Tonsille im immunologischen Geschehen. *Arch Klin Exp Ohr Nas Kehlkopfheilk* 196 65
- Mauget, M. 1959 Modifications de la technique de Charpy en (traçage de l'osmium-iodure de potassium. Résultat de son application à l'étude des fibres nerveuses. *C R Soc Biol (Par.)* 153 939
- , et zum Gontenberge A. & Koburg, E. 1967 Die Tonsille als lymphatisches Organ. *Acta Otolaryng (Stockh.)* 63 79
- Mishima, Y. 1966 Melanosomes in phagocytic vacuoles in Langerhans cells. Electron microscopy of keratin-clipped human epidermis. *J Cell Biol* 30 417
- Mootz, W., Schondorf, J. & Mautz, E. 1971 Langerhans-Zellen im Tonsillenepithel. *Arch Klin Exp Ohr Nas Kehlkopfheilk* 199 604
- Mootz, W., Mautz, E. & Schondorf, J. 1972. Elektronenmikroskopische Nachweise von Langerhans-Zellen im Karyonepithel der menschlichen Gaumenmandel. *J Mund-Gesichtschir* 47 11
- Moser, F. & Braun, H. 1964 Die bindungsbedingte Indurierung in den Tonsillen und ihre pathologischen Zusammenhänge mit den Lymphgewebefortoren. Vergleichende klinische und pathologisch-anatomische Untersuchungen an Tonsillen Rheumakranke. *Arch Ohr Nas Kehlkopfheilk* 164 379
- Nagmann, H. H. 1964 Fluoreszenzmikroskopische Untersuchungen zur Frage der Tonsillenfunktion. I Mitteilung: Das Blutgefäßsystem der Gaumenmandel. *Z Laryng Rhinol Otol* 33 359
- 1957 Fluoreszenzmikroskopische Untersuchungen zur Frage der Tonsillenfunktion. 5. Mitteilung: Retikuloendotheliales System, Plasmazellen, Lymphocyten und Antikörperbildung. *Z Laryng Rhinol Otol* 36 195
- 1970. Diskussionsbemerkung. In: *Arch Klin Exp Ohr Nas Kehlkopfheilk* 196 103
- Nosaka, Y. 1966. Über die elektronenmikroskopische Untersuchung der Tonsillen und einige Folgebefunde (mit Film). *Misch Otorheilk* 100 34
- Reynolds, E. S. 1963 The use of lead citrate at pH 10 as an electronopaque stain in electron microscopy. *J Cell Biol* 17 208
- Sagebiel, R. W. & Reed, T. H. 1968 Serial reconstruction of the characteristic granule of the Langerhans-cell. *J Cell Biol* 36 495
- Schätzle, W. & Falk, P. 1962. Der Einfluß von Nebennierenrindenhormonen auf die postnatale Tonsillenentwicklung beim Kanarienvogel. *Arch Ohr Nas Kehlkopfheilk* 179 227
- Schroeder, H. E. & Theilade, J. 1966 Electron microscopy of normal human gingival epithelium. *J Periodontol* 1 95
- Schwarz, M. 1933 Form und Funktion der Tonsillen. *Z Laryng Rhinol Otol* 32 11
- Specht, W. 1970. Färben mit Aldehydchlorid. Eine Methode für den topochemischen Nachweis von Sulfonacuren und Aldehyden. 63. Mitt. *Ann Ges. Würzburg*
- Süß, P. 188... Zur Physiologie der Tonsillen. *Bd Zbl* 2 368
- 1884 Über Mandeln und Balddrüsen. *Arch Path Anat* 97 211
- Watson, M. L. 1958 Staining of tissue sections for electron microscopy with heavy metals. *J Biophys. Biochem Cytol* 4 475
- Watzka, M. 193... Epithel und Lymphocyte. *Ann Anat* 75 150
- Woff, K. 1967 a. Die Langerhans-Zelle. Ergebnisse neuerer experimenteller Untersuchungen. *Arch Klin Exp Derm* 29 34
- 1967 b. The fine structure of the Langerhans-cell granule. *J Cell Biol* 35 468
- Yamaguchi, Y. 1960 Histopathological studies on the subepithelial basement membrane of the palatine tonsils. *J Otorhinolaryng Soc Jap* 61 (suppl. 1) 11 (auch *Zbl Hals Nas Ohrenheilk* 69 199 (1960 61))
- Zelickson, A. S. 1966 Granule formation in the Langerhans-cell. *J Invest Derm* 47 498

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DISCUSSION

Friedmann: Our observations have been identical and confirm the previous migration of lymphocytes into and through the surface epithelium. The most interesting phenomenon and raises several questions. Are these lymphocytes a waste product of lymphoid hyperplasia and are they shed passively?

the tonsillar crypts or onto its surface. There are often small capillaries opening into the surface epithelium which may carry normal or damaged cells. I am, however not entirely happy with the term "Retikulierung" which might lead to some confusion, at any rate, in the minds of some pathologists. The histopathology of the tonsil presents difficulties and the terminology should be changed and would add to the great value of these investigations.

C. R. Pfaltz: Welche Rolle spielen die Lymphocyten, welche sich während der Retikulierungsphase innerhalb der kryptennahen Epithelschicht befinden? Wo finden nach der Meinung von Herrn Falk die Antigenaufnahmungsprozesse in der Tonsille statt?

P. Falk (Antwort) an Herrn Pfaltz: Wir verbinden mit der Retikulierung einen anderen Begriff. Unter normaler physiologischer Retikulierung des Tonsillenepithels verstehen wir die folgenden verschiedenen Zustandsbilder am Epithel: Die Retikulierung beginnt mit einer wellenförmig verlaufenden Basalmembran, Lymphocyten liegen unter der eben beschriebenen Basalmembran. Zwischen Epithelzellen entstehen Lücken in der Basalmembran, durch welche die Lymphocyten eindringen. Die Epithelzellen geben

durch weitere Lymphocyten auseinander so daß Lymphocyten und Plasmazellen in einem Odemsee zwischen den auseinandergezogenen Fortsätzen der Epithelzellen bis zum Stratum superficiale liegen. Dieses Bild ohne Eindringen oder Darzweckenliegen von Kapillaren oder Bindegewebsfasern nennen wir „physiologische Retikulierung“. Wachsen infolge einer granulierenden Entzündung bei chronischer Tonsillitis durch die Lücken der Basalmembran zwischen den Epithelzellen Bindegewebsfasern und Kapillaren ein, dann können wir von einer „pathologischen Retikulierung“ sprechen, die wir aber bei unseren Untersuchungen nicht feststellen konnten.

An Herrn Pfaltz: Da man im retikulierten Epithel neben Lymphocyten auch Lymphoblasten, Lymphoblasten in Teilung und Plasmazellen findet, muß man annehmen, daß schon im Epithel immunologische Vorgänge stattfinden. Die Lymphocyten können als Träger zellständiger Antikörper in das Tonsillengewebe zurückkehren, wo dann die weiteren immunologischen Vorgänge sich abspielen können. Das retikulierte Epithel als eine Kontaktstelle ist unseres Erachtens ein funktionell wichtiges Glied in einem miteinander verbundenen Immunsystem.

POLYCHONDRITE CHRONIQUE ATROPHIANTE

Ultrastructure des Lésions Cartilagineuses au Niveau du Pavillon de l'Oreille

R. Grimaud et B. Bodelet

Travail de la Clinique O.R.L. de l'Hôpital de Nancy de la Clinique O.R.L. de l'Hôpital de Saint-Dié et du Laboratoire de Microscopie Electronique de Nancy Nancy France

Abstract Après une étude ultrastructurale du cartilage auriculaire humain, les auteurs envisagent les différentes altérations rencontrées chez un malade atteint de polychondrite chronique atrophiante. Si certaines plaques conservent la structure classique du cartilage auriculaire, on note une importante dédifférenciation des chondrocytes au niveau des zones atteintes. Cette dédifférenciation se traduit par une perte de la charge glycogénique et par l'appétence de plus en plus grande du chondrocyte devenu fibroblaste à élaborer des fibres collagènes. La polychondrite chronique atrophiante peut donc être envisagée comme une maladie due à une dédifférenciation progressive des chondrocytes en fibroblastes.

Décrite la première fois par Jahsch-Wartenhorst en 1923 la polychondrite chronique atrophiante reste une maladie rare, mal connue, et d'évolution fatale. Elle rentre dans le vaste cadre des maladies dites « de système » puisque elle atteint aussi bien les cartilages auriculaires, que les cartilages articulaires, bronchiques ou septaux.

Les différents tableaux cliniques de cette maladie sont maintenant bien connus, mais il faut reconnaître qu'ils ne permettent à aucun moment, d'affirmer la polychondrite chronique atrophiante. Seule la biopsie cartilagineuse permet de poser avec le maximum de certitude, ce diagnostic.

Il ne semble pas, de l'avis des auteurs, qu'il existe une chronologie quelconque dans l'atteinte des cartilages, et l'on pourrait tout aussi bien biopsier les anneaux trachéaux (atteints

dans 70 % des cas selon Dolan) que le cartilage de la cloison (déformé dans 82 % des cas) ou encore que le pavillon de l'oreille (tuméfié dans 88 % des cas). En réalité, ce sera le plus souvent au niveau du cartilage du pavillon que l'on fera ce prélèvement. En effet, à ce niveau, il existe généralement des formations cliniquement faciles à dépister et la biopsie sera moins dangereuse qu'au niveau de l'arbre trachéo-bronchique ou des cartilages épiphysaires.

Les lésions histologiques ont été bien décrites en microscopie optique, elles n'ont toutefois pas permis d'apporter de nouveaux arguments de discussion quant à la pathogénie de l'affection. C'est la raison pour laquelle nous avons entrepris une étude en microscopie électronique.

La structure du cartilage auriculaire d'un sujet atteint de polychondrite chronique atrophiante est très nettement dystrophique. Les altérations sont visibles au niveau de la substance fondamentale et au niveau des chondrocytes.

La substance fondamentale

La substance fondamentale du cartilage auriculaire de l'homme comprend deux zones (Fig. 1)

une zone péricellulaire composée de fines

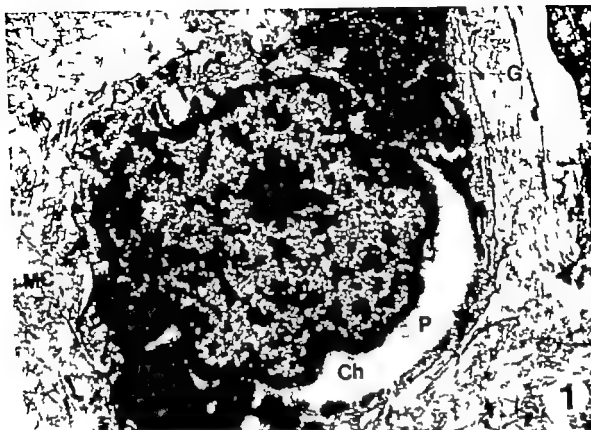


Fig. 1 Cartilage auriculaire humain-polychondrite chronique atrophiante. Le chondrocyte (Ch) garde sa charge glycogénique (G) alors qu'apparaît très nette-

ment le réseau de protofibrilles collagéniques (P). Noter la densification de la matrice (A/C). 2 640.

illes correspondant peut être à la « capsule » des classiques et qui forme la « matrice sombre ».

un réseau de fibres plus épaisses, noyées dans une substance amorphe et qui constitue pour « anglo-saxons, la substance « inter-territoriale » encore appelée « matrice sombre ».

Le cartilage auriculaire d'un sujet atteint de polychondrite chronique atrophiante se pré-

sente au contraire comme un vaste réseau ordonné de fibres collagènes, où il est impossible de reconnaître la matrice sombre et matrice claire. Ce collagène, très dense

(Fig. 2, 3) est parsemé de « plaques » de substance « nuageuse » identique à celle que l'on trouve au sein de la substance inter-territoriale normale.

Les chondrocytes

Il est difficile de parler encore de chondrocytes, tant les cellules observées semblent en avoir perdu peu à peu les attributs.

Leur forme, grossièrement ovale, rappellerait volontiers celle des chondrocytes, mais leur pourtour est beaucoup moins dentelé. Les prolongements cytoplasmiques sont en effet moins nombreux, et ils vont diminuer parallèlement au degré de différenciation des chondrocytes. Le noyau des cellules contient encore, comme pour un chondrocyte normal, un nucléole bien visible.

C'est au niveau du cytoplasme que siègent, en fait, les modifications les plus frappantes.

Tout d'abord, on note une diminution de la charge glycogénique des cellules qui va s'accom-

POLYCHONDRITE CHRONIQUE ATROPHIANTE

Ultrastructure des Lésions Cartilagineuses au Niveau du Pavillon de l'Oreille

R. Grimaud et B. Bodelet

Travail de la Clinique O.R.L. de l'Hôpital de Nancy de la Clinique O.R.L. de l'Hôpital de Saint-Dié et du Laboratoire de Microscopie Electronique de Nancy Nancy France

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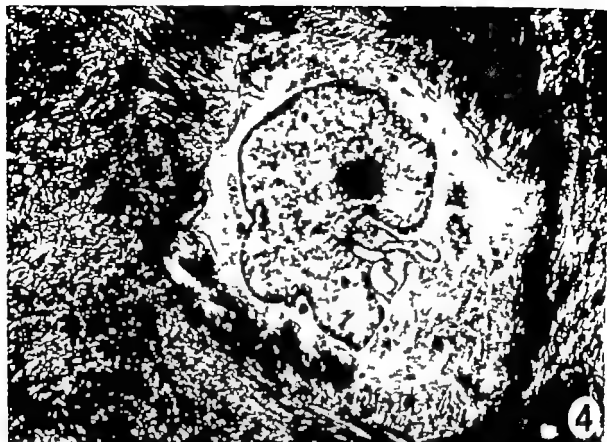
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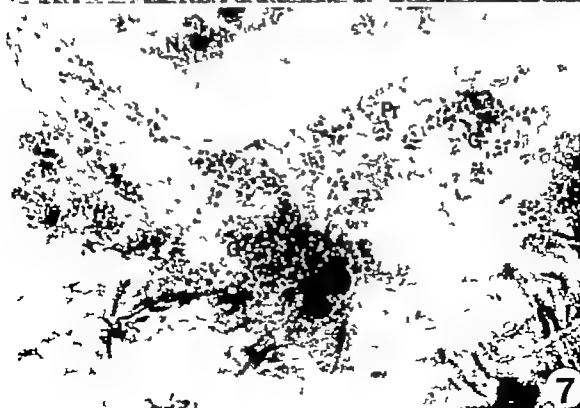
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une zone péricellulaire composée de fibres









Kollagenfasern zu entwickeln. Die chronisch-atrophische Polychondritis kann somit als eine Krankheit bezeichnet werden.

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DISCUSSION

M. Arsen. L'auteur montre deux cas de polychondrite atrophique chez lesquels l'étude ultramicroscopique de la cartilage confirme les résultats de Mr Grimaud. Selon la pensée de l'auteur ce que les pathologues allemands appellent « maladie de Askanazy » est une collagénose dans le domaine O.R.L.

R. Grimaud. (Réponse) Il Mr Arsen: Il est tout à fait justifié de faire rentrer la P.C.A. dans le cadre des maladies mésochymateuses car il existe des atteintes tégumentaires associées dans à des altérations du derme, altérations portant sur la structure fibrillaire.

VAGAL AFFERENT INFLUENCES ON THE PHASIC ACTIVITY OF THE POSTERIOR CRICOARYTENOID MUSCLE

H. Fukuda C. T. Sasaki and J. A. Kirchner

*From the Section of Otolaryngology Department of Surgery
Yale University School of Medicine New Haven Conn., USA*

Abstract The respiratory activity of the posterior cricoarytenoid muscle is strongly influenced by both pulmonary stretch and pressor receptors. These influences are mediated by afferent fibers in the vagus nerves. This demonstration is made possible by the unique anatomic arrangement of the right vagus nerve in the cat, which allows accurate separation of afferent from efferent fibers in its cervical portion.

The primary role of the larynx from a phylogenetic point of view is sphincteric, protecting the lower airway from the intrusion of foreign bodies. In higher species the larynx plays a more definitive role in respiration by assuming a dilator function as well. This is evidenced by rhythmic motor nerve discharges in the laryngeal nerve (RLN) synchronous with the inspiratory phase of respiration (Joels Samueloff 1956 Bianconi & Raschi, 1964)

Green & Neil (1955) from their observations of impulse activity in the RLN and electromyographic activity in the intrinsic laryngeal muscles, concluded that the abductor muscles contracted during inspiration, the adductor muscles during expiration. Similar findings were demonstrated in human beings by Faaborg Andersen (1957).

Pressman & Kellman (1955) investigated the respiratory movements of the vocal cords in man using high speed cinematography and showed that the glottis indeed widened during inspiration and narrowed during expiration.

This work was supported by USPHS - RO 1-NS-05465

According to Negus (1949) the widening of the glottis preceded a contraction of the diaphragm whereas glottic narrowing heralded the onset of expiration. Murakami & Kirchner (1972) however demonstrated that adduction during expiration was not caused by activation of the adductor muscles but by a gradual fall in tonic activity of the posterior cricoarytenoid muscle (PCA)

In further support of the respiratory function of the larynx, it has been demonstrated that under certain circumstances vocal cord movements continue after tracheotomy and that vocal cord abduction precedes any activity in other respiratory muscles (Nakamura et al. 1958).

In this regard, it is generally agreed that the origin of spontaneous rhythmic respiratory activity is located within the medulla, and that this activity is complexly modified by extramedullary nervous influences. These include, among others, the effects of discharges from the pons and reflex impulses ascending in the vagus.

Several observations support this idea. Herling (1868) and Breuer (1868) observed that lung inflation inhibited the inspiratory phase of respiration. Head (1889) on the other hand showed that lung deflation resulted in strong inspiratory excitation and that this phenomenon was abolished by freezing the vagus nerve. Widdicombe (1954) reported similar findings

with cooling of the vagus nerves. Moreover electrical stimulation of the superior laryngeal nerve (SLN) in some circumstances inhibits inspiration (Mårtensson 1963).

With specific regard to the respiratory function of the larynx, evidence suggests that the activity of the PCA is influenced by variations in oxygen and carbon dioxide tension, by anesthetic levels and by any stimulation of the larynx or neighboring pharynx (Suzuki & Kirchner 1969).

Variations in intrapulmonary pressures are also important, as evidenced by the decline in phasic activity of the PCA as ventilatory resistance decreases (Sasaki et al. 1972).

These observations therefore suggest that the respiratory function of the larynx originates in the medullary respiratory center and that this respiratory center may be influenced by afferent stimuli of extramedullary origin, chiefly those mediated by the vagus nerves.

It is therefore our purpose to clarify the effects of vagal afferents on laryngeal respiratory function with special regard to the effect of pulmonary stretch and pressor afferents on phasic abductor activity.

METHODS

Thirty-one healthy adult cats were used in this experiment. Anesthesia consisted of sodium pentobarbital, 30 mg/kg body weight administered intraperitoneally. A uniform degree of anesthesia was maintained throughout the experiment by intravenous augmentation at intervals as necessary.

The larynx and trachea were exposed through a vertical midline incision. The strap muscles were then identified and completely removed. The upper half of the sternum was partially resected to obtain further exposure of the superior mediastinum, care being taken avoid injury to the pleura.

The trachea was then transected at the lower margin of the larynx; the distal segment cannulated and fitted with a three-way

stopcock to facilitate deliberate inflation and deflation of the lung.

The following nerves were then exposed, carefully avoiding injury:

- 1) right and left SLNs
- 2) right and left VAGI
- 3) RLNs (descending)
- 4) RLNs (ascending)

Our plan was to isolate both descending RLNs at their take-off from the vagus, so as to preserve the motor nerve supply to the larynx while observing the effects of blocking or dividing the vagus just below this point. However the left RLN could not be exposed at its point of origin from the vagus without producing pneumothorax. On the right side, the point of origin was easily identified, the subclavian vein providing a convenient landmark. The RLN usually lies deep to this vein within the carotid sheath at the level of the 15th to 17th tracheal rings (Fig. 1).

Complete vagal de-afferentation was accomplished by blocking or dividing the right vagus below the RLN take-off and the left vagus in the neck. Since the motor supply to the larynx was spared only on the right side the right PCA muscle was used for recording.

In order to eliminate other minor afferents, both SLNs were cut prior to the experiment. Since there is some evidence that the sympathetic system conducts afferent impulses (Cromer et al., 1933), the sympathetic trunks were also sectioned in the neck.

Concentric needle electrodes were used, the EMG being displayed on a multichannel oscilloscope (Tektronix 564 B) with simultaneous amplification on the Grass AM5 sound system.

Intratracheal pressure was measured by a Statham P23BC transducer and displayed in phase on the Tektronix multichannel oscilloscope.

In some instances, the peripheral segment of one vagus nerve was divided into single fibers or filaments which were positioned on platinum wire electrodes within a warm mineral oil bath. Neural activity was then displayed

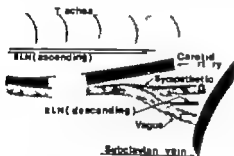


Fig. 1 The anatomy of the right vagus in the neck allows selective separation of afferent fibers from efferent (Recurrent Laryngeal Nerve).

oscilloscopically after amplification by a DAM 5 differential preamplifier. All oscilloscopic activities were photographed by the Grass camera.

Inflation of the lung was carried out by forcing 30 ml of air directly into the three way stopcock. Deflation was carried out by aspirating through the same stopcock before the onset of spontaneous inspiration.

RESULTS

Inflation of the lung

With both vagi intact, the activity of the PCA muscle during inspiration was normally re-

duced or abolished by inflation of the lungs. On the other hand, when both vagi were divided as described above, lung inflation caused no discernible effect on the existing level of spontaneous activity in the right PCA. It is important to point out that bilateral vagotomy generally resulted in an increased level of inspiratory activity presumably because of interruption of vagal inhibitory fibers. It is upon this level of spontaneous PCA activity that lung inflation had no discernible effect.

Deflation of the lung

With intact vagi, deflation of the lungs normally resulted in accentuation of PCA activity

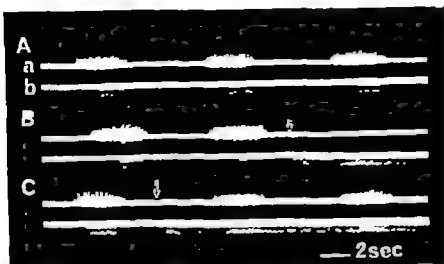


Fig. 2 Vagal afferent influences on inspiratory activity of the posterior cricoarytenoid muscle. (a) (Top line in all three tracings). Electromyogram of the right posterior cricoarytenoid muscle. (b) (Lower line in all three tracings). Neural potentials in afferent filament in right vagus. *A* Recording during normal respiration. *B* At the arrow the lung was mechanically in-

flated. Posterior cricoarytenoid activity stopped at once, and afferent vagal potentials started, continuing as long as the lung remained inflated. *C* Same procedure as *B* but after additional section of the left vagus in the neck. Inflation of the lung induced afferent neural potentials, but the right posterior cricoarytenoid muscle continued its phasic activity

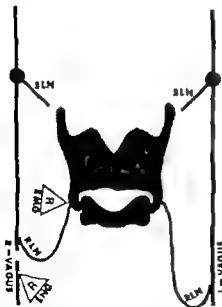


Fig. 3 This figure shows that the pathway of the vagal afferent impulses which inhibit the posterior cricoarytenoid activity in Fig. 2 is the left vagus and that the vagal afferents were recorded from the peripheral segment of the right vagus. *R* Recording electrode. *ENG* Electroneurogram. Both superior laryngeal plexus and both sympathetic trunks have been cut in this experiment and in Fig. 5

during inspiration. Deflation caused an increase in both duration and frequency of PCA discharges.

After both vagi were sectioned as described above, the level of spontaneous activity appeared unaffected by forced lung deflation.

Increased ventilatory resistance

With the vagi intact, increasing airway obstruction caused strong activity of the PCA muscle and simultaneously induced high negative intratracheal pressures.

After bilateral vagotomy as described above, the level of spontaneous activity in the intact PCA again appeared unaffected by airway obstruction.

PCA activity and afferents through the vagus

Figs. 2 and 4 show simultaneous electromyograms of the right PCA muscle and the afferent electroneurograms of a single fiber preparation of the vagus (Fig. 2, right vagus, Fig. 4 left vagus).

When the lung was mechanically inflated at the end of a normal inspiratory effort, dis-

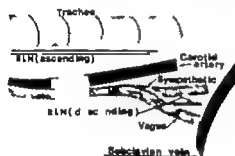


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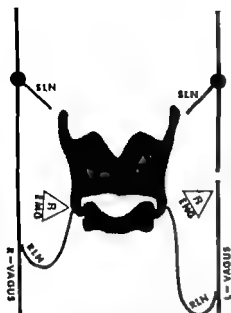


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RESUME

L'activité respiratoire du muscle crico-aryténoïdien postérieur est fortement influencée par les récepteurs pulmonaires d'étirement et de pression. Les influxes provenant de ces récepteurs sont conduits par les fibres afférentes du nerf vague. La démonstration de cette action est facilitée par la disposition anatomique spéciale du nerf vague droit chez le chat; celle-ci permet, en effet, dans la portion cervicale, de séparer précisément les fibres afférentes des fibres éférentes.

ZUSAMMENFASSUNG

Die Atmungsfunktion der hinteren Cricohyarytenoid-Muskeln wird sowohl durch die Lungenspannung als auch durch die Pressor-Rezeptoren stark beeinflusst. Diese Einwirkungen werden durch zuführende Fasern des Vagus-Nerven vermittelt. Dies kann wegen der ungewöhnlichen anatomischen Struktur als rechten Vagus-Nerven der Katze demonstriert werden, an dem es möglich ist, zuführende und abführende Fasern in seinem Halsabschnitt genau zu trennen.

REFERENCES

- abolishes the inspiratory activity of the PCA muscle.
 - 2) Forced deflation of the lung results in increased inspiratory activity of the PCA muscle.
 - 3) After vagal de-afferentation, neither inflation nor deflation affects the level of spontaneous respiratory activity in the PCA muscle.
 - 4) Increased ventilatory resistance which presumably stimulates pulmonary pressor receptors induces strong PCA muscle activity
 - 5) After bilateral vagal de-afferentation, increased ventilatory resistance fails to influence spontaneous PCA muscle activity
- This study demonstrates that the respiratory activity of the PCA muscle is strongly influenced by both pulmonary stretch and pressor receptors and that these influences are in fact mediated by afferent fibers in the vagus nerves. This demonstration is made possible by the unique anatomic arrangement of the feline vagus which allows accurate separation

- Bianco, R. & Raschi, F. 1964 Respiratory control of motoneurons of the recurrent laryngeal nerve and hypoglossal space. *Arch Ital Biol* 102 56.
- Brewer J 1868 Die Selbststeuerung der Atmung durch den Nerven Vagus. *Sitzber Akad Wiss Wien* 58 909
- Cromer S. P. Young, R. H. & Ivy A. C. 1933 On the existence of afferent respiratory impulses mediated by the stellate ganglia. *Amer J Physiol* 104 468.
- Faaborg-Andersen, K. 1957 Electromyographic investigation of intrinsic laryngeal muscles in humans. *Acta Physiol Scand* 41 Suppl. 140
- Fukuda, H. & Kirschner J. A. 1972 Changes in the respiratory activity of the cricothyroid muscle with intrathoracic interruption of the vagus nerve. *Ann Ctol*. In press.
- Green, J. H. & Neill, E. 1955 The respiratory function of the laryngeal muscles. *J Physiol* 129 134
- Head, H. 1889 On the regulation of respiration. Part 1 Experimental. *J Physiol* 10 1 Part 2, Theoretical. *J Physiol* 10 279
- Herrig, E. 1868 Die Selbststeuerung der Atmung durch den Nerven Vagus. *Sitzber Akad Wiss Wien* 57 672.
- Joch, N. & Sammeloff, M. 1956 The activity of the medullary centers in diffusion respiration. *J Physiol* 133 360.
- Krowton, H. C. & Larrabee, M. G. 1946 A preliminary analysis of pulmonary volume receptors. *Amer J*

- Larrabee, M. E. & Knowlton, G. C. 1946. Excitation and inhibition of phrenic motoneurons by inflation of the lungs. *Amer J Physiol* 147: 90.
- Mårtensson, A. 1963. Reflex responses and recurrent discharges evoked by stimulation of laryngeal nerves. *Acta Physiol Scand* 57: 248.
- Murakami, Y. & Kirchner J. A. 1972. Respiratory movement of the vocal cords. An electromyographic study in the cat. *Laryngoscope* 82: 454.
- Nakamura, F., Uyeda, Y. & Sonoda, Y. 1958. Electromyographic study on the respiratory movements of the intrinsic laryngeal muscles. *Laryngoscope* 68: 109.
- Negus, V. E. 1949. *The comparative anatomy of the larynx*. Heinemann, London.
- Pressman, J. J. & Kelman, E. 1955. Physiology of the Larynx. *Physiol Rev* 35: 306.
- Sasaki, C. T., Fukuda, H. & Kirchner J. A. 1972. Laryngeal abductor activity in response to varying ventilatory resistance. In press.
- Suzuki, M. & Kirchner J. A. 1969. The posterior cricoarytenoid as an inspiratory muscle. *Ann Otol* 78: 849.
- Widdicombe, J. G. 1954. Respiratory reflexes excited by inflation of the lung. *J Physiol* 123: 105.

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DISCUSSION

I. Padoyev. I have two questions from a more technical point of view.

1. Why did Mr Kirchner use in his experiments the cat and not the dog? It is well known that the anatomy of respiration is not the same and the one in a dog is more like that of man, especially the yuv. Since I am sure the difference in price between a cat and a dog in the USA cannot cause a problem there must be another reason for his choice.

2. It would be of interest to know if Mr Kirchner had in mind the investigations by Felberbaum in Leningrad and also those by the group from Mount Sinai Hospital in New York dealing with receptors

of the respiratory pathways and especially of the larynx.

E. Loebell. Did Mr Kirchner find different kind of spindles? And were the studies done in relation to the inspiratory airflow? In my electromyographic studies of the diaphragm I found two types of inspiration in humans: most start inspiration with electrical activity of the diaphragm. In other cases this activity starts later and lasts longer than the respiratory curve of inspiration.

J. A. Kirchner (Reply) to Mr Padoyev. The cat is the best animal for experimental neurophysiology for many reasons, including the fact that their nerve trunks contain very little fibrous tissue, so that nerve fibers can be easily identified, anatomically and physiologically. And they are also less expensive than dogs!

Mechanoreceptors in the larynx have been studied by many investigators. Those located in the oses have been well described by König of Heidelberg, and von Leden of Los Angeles. Those located in the fibrous capsules of the laryngeal joints have been identified morphologically by two Russians, Inkavskaya and Gracheva, and studied physiologically by Wyke and his group in London.

The evidence for muscle spindles is less certain, and varies among animal species. Spiral endings have been photographed by Wyke and Abo-el-Easa, but whether these structures act as genuine spindles is not certain. Mårtensson, for example, has not found physiological evidence for spindles in the laryngeal muscles of the animals he studied.

Nevertheless, the corpuscular nerve endings in the laryngeal tissues undoubtedly serve a protective-reflex function, and may even be important in a feed-back control of the laryngeal muscles during phonation.

To Mr Loebell. The posterior cricoarytenoid muscle begins its contraction before the diaphragm, and appears to be influenced by the same changes in pO_2 , pCO_2 , intrapulmonary pressure, etc.

Regarding your second question, we have observed two types of muscle fibers—or better—motor units in the posterior cricoarytenoid muscle: one type shows the phasic, inspiratory activity and serves to open the glottic. The other type does not show this phasic activity and may have a phonatory function in stabilizing the arytenoid cartilage against the forward pull of the thyro-arytenoid muscle. But I cannot be sure

NASOPHARYNGEAL FIBROMA

R. Tapia Acuña

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Abstract The purpose of this paper is to present my experience on juvenile nasopharyngeal fibroma, a disease which is tremendously frequent in Mexico. We have done some research on its etiology from the points of view of genetics, heredity virus infection, and on treatment.

Since 1956 when I reported 70 cases of Juvenile Nasopharyngeal Fibroma (Tapia Acuña, 1956), many more cases have been seen at the General Hospital and in private practice in Mexico City making a total of 279 within the last 22 years. We saw 15 cases in the first half of 1970 alone, resembling an epidemic disease. The patients came from different altitudes and latitudes, including big towns. This fact caused me believe that a virus and not a hormonal factor might be responsible for it, which has been previously investigated by us and some others.

In all cases here reported, diagnosis was made clinically and confirmed by histopathological examination, which demonstrated the presence of stellate cells supported by immature fibroblasts with tortuous vessels in a stroma of myxomatous tissue, devoid of elastic or muscular fibers (Figs. 1-3).

Clinical diagnosis was based on the following symptoms and signs: nasal obstruction, repeated hemorrhages from the nose and pharynx and presence of a hard tumor situated at the nasopharynx, usually involving one or both nasal fossae and one or more facial sinuses and sometimes the orbit or the pterygomaxillary space. The sella turcica or even the brain were involved in a few cases. Ear troubles and

infection were sometimes present, pain was rare and the general condition was fair except when the patient had suffered abundant or very frequent hemorrhages. Facial deformities and unilateral exophthalmus with vision troubles (Figs. 4-5) were not rare in our cases. During clinical examination, the tumor was seen to be smooth, grey or reddish, with a tendency to bleed at any contact, manual or instrumental (Fig. 6).

Radiological findings were: presence of a shadow occupying the nasopharyngeal space and one or more sinuses, deformity of the sella turcica, etc., all depending on the elongations and size of the tumor. Tomography confirmed the above-mentioned data. Angiography showed vessels surrounding or penetrating into the tumor in a definite way.

An examination of the etiology of this disease was started with the purpose of discovering the possible influence of a genetic defect or a virus infection in the development of the tumor. Thirty-five patients were studied by the geneticist and some fresh specimens were sent to the virologist to look for unknown virus.

I may sum up the results of this study and our own clinical experience as follows:

(a) The ages of the patients ranged from 10 to 25 years. Recently I saw a patient 35 years old having a large nasopharyngeal fibroma. He had been operated at the age of 17 years for the same complaint, without improvement, which explained the persistence of the disease.

(b) We have never seen a case of juvenile nasopharyngeal fibroma in females. In all cases



Fig 1 Hyaline fibrous tissue. Appressed vessels. Limit of the tumor at level 36.

studied, the patients were male, having normal genital organs and the XY chromosome pair

(c) There is no relation between heredity and the development of the tumor. All examined patients had a normal karyotype and no abnormality or genetic disease was found either in them or in their relatives.

(d) Most of the patients were Mexican natives. The remainder were mustee or of European extraction.

(e) No specific infection has been found.

Only common flora of the respiratory tract and a few adenovirus were present in the specimens sent for virus study.

(f) Slight increase in white blood cells and decrease in red cells and hemoglobin was the only pathologic finding in blood tests, though not in all cases.

(g) Biopsy is unnecessary if a correct clinical diagnosis has been established. Sometimes biopsy could be dangerous because of the possibility of causing a severe hemorrhage in



Fig 2 Fibroblastic myxomatous tissue and vessels. No elastic fibers. 36

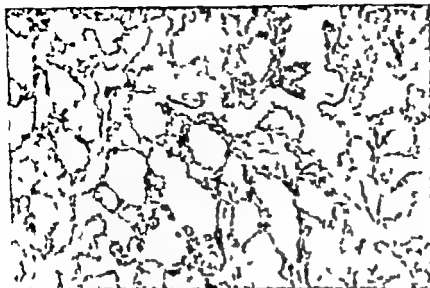


Fig 3 Multiple vascular canals. 36

might impair the patient's general condition or the surgical treatment more difficult.

Medical treatment is useful only to prepare patient for surgery but will not cure him. Radium, X ray and cobalt therapy have failed in our hands. Electrocoagulation might be of help in cases of small or residual tumors, though results are not always satisfactory. Hormones or any other hormonal administration not only do not help, but produce undesirable side effects. In addition, I have tried electrocoagulation to assess its destructive action on specimens of this kind, though without positive results.

Surgery remains the method of choice. Usually we do it when the general condition of the patient is at its best. Nevertheless, removal of the tumor has sometimes been considered urgent due to massive or too frequent hemorrhages or when its size is compromising vision or other function. We have observed that small, better than large blood transfusions a few days before operation are helpful. During operation, we transfuse 1 000 cc of blood. Intubation for anesthesia through a tracheal tomo instead of through the mouth, is preferred when the tumor is too large.

The surgeon has to bear in mind three problems, which are: a proper approach, a

complete removal of the tumor and control of hemorrhage during operation. The usual way is the retropalatine approach. I perform a horizontal incision on the soft palate. The tumor is then seen through this window. The surgeon's finger is introduced through the incision to palpate the tumor, assess its size, extension, point of origin, adhesences and everything which is useful for a good dissection. After pulling out and upward the soft palate, the tumor is held with a proper forceps, the finger is replaced into the opening and will serve to guide a dissector or curette, which is introduced via one nostril in order to make



Fig 4 Facial deformity. Fibroma was removed by the combined oral and retropalatine approaches.



Fig 5 Exophthalmos. The deviated eye returned to its place after removal of the tumor

the dissection. An assistant pulls the forceps downward and once the tumor has been isolated, it comes out without difficulty. The procedure has to be done rapidly to avoid excessive blood loss. Hemostasis by packing is the following step.

In some cases it was necessary to perform later rhinotomy with or without partial removal of the skeleton, combining this approach with the one mentioned above. I had to

open through the mouth the pterygomaxillary space four times and once to open it through the maxillary sinus. On two occasions I did not dare to remove the whole tumor because it had extended too far into the cranial fossa. The most amazing case I have yet seen is that of a boy 12 years old who had part of the tumor inside the brain (Figs. 7-8), without disturbing his vision or causing troubles other than those commonly seen. I was able to reach the opening of the cranial fossa and I cut the tumor at this level. An expert neurosurgeon performed temporal lobectomy but he could not remove the remaining tumor because he found difficulty in dissecting it and controlling the hemorrhage. This patient left the hospital apparently in good condition. He came to the otolaryngological department one year later with a slight difficulty in walking. His nose and nasopharynx were free from tumor reflexes were normal as well as his psychological sphere. He refused a new examination, which would have been of interest.

In our series we have to regret just one fatality which occurred 17 hours after removal of the fibroma by lateral rhinotomy. The



Fig 6 Specimen: the middle fragment was located at the sphenoidal sinus; fragment at left was at the pterygomaxillary fossa.



Figs 7 and 8 Intracranial nasopharyngeal fibroma.

surgeon could not convince the parents of the patient to authorize autopsy. Recidives or residual tumors have been observed in 25 cases (9%). Most of the patients were operated on. Others were treated by electrocoagulation.

In conclusion I believe that the frequency of cases is related to race and nutrition. Surgery is the method of choice for treatment. Cryosurgery as used by Miller (1969) and there might be a good solution to the problem hemorrhage. I am satisfied with the results of our procedures, though I think that there are still ways of improving them. Work has to be done in the field of immunology for prevention of the disease, which is impossible, so far.

RÉSUMÉ

but de ce travail est de faire connaître notre expérience sur le nasofibrome-juvénile dont la fréquence

est extraordinaire au Mexique. Nous avons fait des recherches du point de vue causal, en tenant compte des facteurs génétiques, héréditaires et viraux. Les résultats du traitement sont analysés.

ZUSAMMENFASSUNG

Der Zweck dieser Arbeit ist, unsere Erfahrungen über das in Mexiko bei jugendlichen außerordentlich verbreitete Nasenrachsenfibrom darzulegen auf Grund von Untersuchungen über die Ursache, wie genetische, erbliche und virale Faktoren.

REFERENCES

- Tapia Acuña, R. 1956. The nasopharyngeal fibroma and its treatment. *Arch Otolaryng* (Chic.) 64: 451.
Miller D. Personal communication. Mexico, August, 1969.

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Fig 1 (a) Angiography of the internal carotid, left side, from a projection. Contrast filling the tumour area.

DISCUSSION

G Hall. In Norway this disease is not as common as it apparently is in Mexico. The treatment also in our country has been extirpation, which, as mentioned, often leads to severe haemorrhage. In one of our cases, this became so severe during the first operation that upon recurrence, we operated in hypothermia and employed extracorporeal circulation.

The patient was a 20-year-old boy suffering from a blocked but running nose for the past 4 years. A tumour in the left side of the epipharynx was discovered in the local hospital, and upon admittance to the State Hospital an angiography revealed that its blood supply came from the left carotid. A hypertrophy of the ascendant palatine and the ascendant and descendant pterygoid arteries was found, and, in addition, a destruction of the dorsal part of the bony palate. The tumour was removed, followed by a severe haemorrhage: three bottles of blood were infused. Following a new haemorrhage 4 days later we had to ligate the external carotid on the left side.

Twenty days after the operation he was discharged, but a new growth of the tumour was seen after only 6 months. The patient had swallowed a good part of the tumour but still the epipharynx was filled up. A new angiography showed an abnormal blood supply from the carotid siphon to the tumour via the ophthalmic artery (Fig. 1 a and b).

Hormonal therapy and X-ray therapy were discussed, but rejected. The patient was able to wait for about 2 years, but then something had to be done and operation in hypothermia and extracorporeal circulation was decided upon.

X-ray tests with compression were performed to see whether the brain had sufficient blood supply from either side alone and also tomometry of the eyes to see whether a compression of the left internal carotid would interfere with the sight of the left eye.

Fortunately these tests showed favourable conditions, and there were no serious contraindications. The operation was performed in March, this year.

The sternum was split, cannulas were introduced through both caval veins, the right femoral artery and



Fig 1 (b) Angiography internal carotid, left side. Contrast filling the tumour area via the ophthalmic artery (→).

the apex of the left ventricle into the heart. The cooling went down to about 16°C, measured in the oesophagus. The total perfusion time was 2 hours and 50 minutes. The approach to the tumour was a combined transpalatal and transpharyngeal one. A part of the hard palate was chiselled out, the tumour removed and a tampon introduced. Six litres of blood were infused, in addition 2 l postoperatively and another litre when the tampons were removed. The condition of the patient was good throughout the intervention, and he was declared to be fit for work after 1 month.

J E Cowley Please describe the technique of using the finger thru the palate and instruments used in this operation and clarify the rate of local recurrence, mortality and inoperable status. And also add a word of caution re the use of silicone spheres in the treatment of these tumors due to the possibilities of intracranial complications.

R Albrecht Dr Acuña has the luck in absolute Häufigkeitsgipfel des Nasenrachenhirns teilig zu sein. Auch in Indonesien und in Teilen des vorderen Orients ist es nicht so selten wie z.B. in Europa. Daher dürfen wir von ihm die Beantwortung von Fragen erhoffen, die an Hand von Einzelfällen nicht recht zu klären sind. So darf ich fragen, ob in Mexico eine familiäre Disposition für das Nasenrachenhirns zu finden ist. Auch die Chromosomenanalyse wäre an einer so grossen Zahl Erkrankter klar angebracht. Bisher wurden bei nur vereinzelt Untersuchungen eine Abweichungen gefunden. Die Frage einer viralen Induktion dieser eigenartigen Geschwulst ist noch nicht definitiv entschieden. Wir haben die elektronen-

optisch überprüft. Dabei fanden wir die eigenartigen, für das Nasenrachenhirns spezifischen Kernschliesskörper von denen wir bisher nur wissen, dass sie nichts mit Viren zu tun haben und dass sie keine Ribonukleinsäure enthalten. Sinn und wirkungsvollster Weg einer möglichen Hormontherapie ist an einem grossen Krankengut natürlich auch am besten bewiesen. Über und ich möchte fragen, welche Erfahrungen hier bereits gemacht wurden. Wir müssen unverändert, wenn auch nur an Hand weniger Fälle bestätigen, dass das Tetraäthylsalz des 4-4-Dihydroxy- α - β -diäthylstilbendiphosphats allen bisherigen Östrogenpräparaten überlegen ist und auch in vitro die Zellteilungswerte des Nasenrachenhirns beeinflusst. Es führt regelmäßig zur Rückbildung der Geschwulst und ist deshalb in besonders problematischen Fällen ein wertvolles Hilfsmittel.

U Sürücü Just a surgical tip: If the nasopharyngeal fibroma is preoperatively filled with solution saline containing few drops of adrenalin and the injection is made with pressure it will anemize the tumour which is then easier to remove.

R Tapia Acuña (Reply) to Mr Hall. I have never seen a case like the one you mentioned. We usually transfuse 800 cm³ to 1 000 cm³ of blood during operation. The worst case I ever saw was that of the boy who had a big part of the tumor inside the brain. This case is already described in the paper. The short time I devoted to present it did not allow me to explain it clearly enough.

to Mr Cowley: I believe that ligation of carotid maxillary artery is unnecessary since bleed operation is approximately the same. The

me to guide the dissector which is introduced through one of the nasal fossae. I have with me a super-8 film showing my surgical technique. I hope Mr Conley will be able to see it sometime.

Only twice could I not remove the tumor totally. This fact is also mentioned in the paper. In my whole series we had to regret the death of one patient operated by somebody else by lateral rhinotomy. Recurrence or residual tumor was observed in 9% of the cases.

to Dr Albrecht: Of course we have tried diethyl

stilbestrol for treatment of the nasopharyngeal carcinoma, but without positive results. On the contrary, it produces undesirable side-effects, as has been shown on one of my slides. It has been demonstrated that there is no genetic or virus factor in the development of the tumor.

to Mr Shirah: I do not agree with your suggestion because it is much better not to touch the tumor unless for its removal. It might bleed or create adhesion, making surgery more difficult.

HOMOTRANSPLANTATION OF CANINE TRACHEA AFTER DENERVATION OF THE SPLEEN

A Preliminary Report

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The rejection phenomenon has been the obstacle to advances in organ transplantation. Immunosuppression has been only partially effective in controlling rejection. The role of the spleen in rejection of transplanted tissues is still but poorly understood. We have performed trachea transplants which are based upon an observation that the rejection phenomenon can be altered through denervation of the spleen. The results of these experiments will be discussed.

In the treatment of tracheal stenosis it might be of advantage if transplanted material could be used. We have therefore made experiments with tracheal homotransplants in dogs. This is a preliminary report of these experiments.

In a symposium on Tissue and Organ Transplantation, writing about the scope of cartilage grafting, Gibson—a senior lecturer in tissue transplantation at Glasgow University—says: "Since allograft cartilage behaves precisely as an autograft there would be little need for the latter." This characteristic is due to the unique properties possessed by cartilage of not provoking an immune response when transplanted heterogeneously. According to Gibson the laryngeal and tracheal cartilages would be ideal for reconstruction after an operative excision in this anatomical area, but he wonders: "How can one create from autogenous sources the mucous lining, the muscles, or the interconnecting fibrous tissue?"

The opinion of Gibson concerning the use of tracheal cartilage as allograft may be considered very optimistic. Alonso et al. (1972) performed homotransplantations experimentally on dogs and found that the 8 and 12 month tracheal specimens (two-thirds of the tracheal circumference) showed complete resorption of the transplant. They stated that "by avoiding the mechanical pitfall of replacing the entire tracheal lumen, partial success with allografts is possible".

It seems that if permanent preservation of the cartilaginous arches in a tracheal segment transplanted as allograft were possible, this would raise hopes of a satisfactory outcome in severe cases of stenosis of the trachea.

Although Gibson maintains that a cartilaginous allograft takes and heals well, it is apparent that the phagocytosis occurring in tracheal cartilaginous arches is a rejection response and its elimination might extend the survival time of these arches.

There are tissues in a transplanted tracheal segment—it is a composite graft—which differ in the ability to provoke immune responses. Cartilage causes the least intense reactions whereas mucous membrane, muscle and other transplanted tissues produce a stronger response.

We decided to test whether the rejection response can be suppressed or eliminated by applying Toivio's new theory of the effect exerted by splenic denervation on this phenomenon.

Toivio has recently put forward a theory according to which the spleen is the centre through which the host's immune response to a transplant is primarily directed, and that immunological information is transferred to the lymphocytes within the spleen by some hitherto unknown neurological mechanism. On the basis of this theory the authors have subjected eight adult dogs to total splenic denervation, but not splenectomy. At the time of denervation and in several dogs later as well, skin homografts from unrelated mongrel dogs have been applied. The authors report that, in every case some or all of the grafts survived without the use of any other form of immunosuppression. At the time of the report the longest survival was 18 months.

This theory sounds surprising and perhaps unbelievable but, if it proves correct, will create quite new possibilities for tissue transplantation.

As mentioned, our experiments with transplantation of canine tracheal homografts were made in an attempt to discover whether the denervated and control dogs differed in homotransplantation acceptance.

According to the literature (Ferguson et al., 1940; Greenberg, 1958; Fleming & Hommerich, 1968; Tala & Mäsmes, 1968; Kornmesser 1970; Alonso et al., 1972) the fate of a homograft of dog's trachea is as follows. Two weeks after transplantation there is hyperemia and edema at the anastomosis. Two to four months after it there is granulation tissue lining the transplanted mucous membrane. The cartilaginous arches of the transplant undergo phagocytosis. This takes place gradually and is completed in 8 to 12 months. Two to four months are required for squamous epithelium to take the place of granulation tissue as lining membrane of the graft. After 4 to 6 months this is again replaced, by ciliated columnar

epithelium with minimal submucosal inflammatory reaction. During this period the surrounding muscle fibres atrophy and undergo hyalin degeneration.

Lymphocytic collections are seen at the site of resorption of the transplanted cartilage and there occurs universal perivascular lymphocytic cuffing throughout the grafted area.

Because of concentric scarring the transplanted segment is shortened and the tracheal wall becomes thinner and the lumen narrower. If the transplanted piece of trachea is too short enough this can result in collapse of the trachea and suffocation of the animal.

According to Kornmesser (1970) and Fleming & Hommerich (1968) the autotransplantations of tracheal segments containing 3 to 5 cartilaginous arches have been successful.

MATERIAL AND METHODS

Our material consists of 16 dogs of which 8 were denervated and 8 were control animals. In each of a pair of dogs a window only was made in the anterior tracheal wall and the excised quadrangular pieces of tracheal wall were interchanged. In all the others a segment consisting of the whole circumference of the trachea enclosing 4-5 tracheal arches and the corresponding membranous part were interchanged. The operation was performed under Hypnostan anesthesia. The dogs were treated with a combination of penicillin and streptomycin postoperatively. They did not get any form of immunosuppression therapy with the exception of the denervation of the spleen.

The condition of the transplant was examined by endoscopy and endoscopic photography, X-ray films and excision and histological examination of the transplanted segment. In 2 dogs the exchange of tracheal segments was made twice. All the dogs survived the operation well.

After being cut off from the supplying blood circulation the transplanted piece of trachea was sutured into place in the new host. It is natural, then, that the parts of the transplant

immediately adjoining the tissue of the host, i.e. the external portions, will fairly soon obtain adequate nutrition. The mucous membrane within the transplant, however is disconnected from the supplying circulation for a longer time. This of course causes damage to the mucosa. The cartilaginous arches and the tissues joining them suffer least injury through the transplantation. The shorter the transplanted segment is, the better are the chances of survival. The final fate depends on the reaction of the host.

The specimens for histological study were obtained by excising the transplanted part of the trachea 8 to 130 days after transplantation. The tracheal stumps were sutured together and the dogs usually survived the procedure.

RESULTS

When considering the possible effect of denervation on the "take" of the transplanted piece of trachea we know from earlier investigations that the transplant gradually disappears and is replaced by tissue growing into the transplant from adjacent structures of the host. If in denervated dogs, we were to find a weakened rejection or permanent acceptance of transplanted tissue, this would be evidence of a changed reaction of the recipient dog.

Several factors seem to have influence upon the fate of the homotransplanted tracheal segment.

Such factors are the width of the transplant compared with the recipient dog's tracheal lumen, the length of the transplant, possible infection or hematoma, the technique of operation, the histocompatibility of the two dogs and the antibiotic therapy. In these circumstances the evaluation of the significance of one single factor such as denervation of the recipient dog's spleen, proved to be very difficult, particularly when the time of observation has not been long enough.

Comparison of the two groups (denervated and control dogs) revealed in both of them, during the first 2-3 weeks after transplanta-

tion, disappearance of the mucous membrane and its replacement by granulation tissue. Gradually this was covered by new epithelium. The growth of this epithelium seemed to be more advanced in the denervated dogs. Forthcoming investigations will probably indicate whether this epithelium originates from the transplanted specimen or from the host animal. There was a marked infiltration by lymphocytes and phagocytosis of the cartilaginous arches. In denervated dogs these rejection phenomena were somewhat less marked. The difference is however just a matter of degree.

The question whether the denervation of the recipient dog's spleen has some influence upon the fate of the tracheal transplant or not—which was the aim of our investigation—cannot yet be fully answered. As mentioned, the immediate rejection response seems to be somewhat weaker in the denervated dogs, and we have two denervated dogs with very good preservation of the cartilaginous arches of the transplanted tracheal segments after 100 and 130 days respectively. None of the control dogs have obtained such a good result. However still more evidence is needed. We are continuing our experiments for the purpose of finding out what is the final fate of homotransplanted tracheal segments in denervated dogs.

It seems that for the elimination of the rejection response of the transplanted tracheal homograft it would be of advantage to combine denervation of the recipient dog's spleen with some other means or agent for immunosuppression.

RESUME

Le phénomène de rejet a constitué le plus grand obstacle au progrès de la transplantation des organes. L'immunodépression a été seulement partiellement efficace à contrôler le rejet. Le rôle de la rate dans le rejet de transplantations est encore très peu connu. On a récemment fait des transplantations de la trachée chez une méthode où le phénomène de rejet est changé grâce à une désinnervation de la rate. Le résultat de ces recherches sera présenté et discuté.

ZUSAMMENFASSUNG

Die Abstoßung des verpflanzten Gewebes ist bisher das größte Hindernis für den Fortschritt der Organtransplantationen gewesen, und mit Hilfe der Immunosuppression konnte die Abstoßungsreaktion nur teilweise beherrscht werden. Welche Rolle die Milz bei der Rejektion des transplantierten Gewebes spielt, ist immer noch mangelhaft bekannt. Wir haben Trachea-Transplantationen beim Hund ausgeführt, die auf der Beobachtung beruhen, dass die Gewebeatstoßung durch Denervierung der Milz verändert werden kann. Die Resultate dieser Experimente werden besprochen.

REFERENCES

- Alonso, W. A., Bridger O. P. & Bordley J. E. 1972. Tracheal transplantation in dogs. *Laryngoscope* 82: 204.
- Ferguson, D. J., Wild, J. J. & Wangenstein, O. H. 1950. Experimental resection of the trachea. *Surgery* 28: 597.
- Fleming, J. & Honnrich, A. W. 1968. Homotransplantation of the trachea in the dog. *Arch. Otolaryngol.* 88: 724.
- Gibson, T. 1967. The transplantation of cartilage. The College of Pathologists. *Tissue and Organ Transplantation*. B.M.A. House, Tavistock Square, London W.C.1. England.
- Greenberg, S. D. 1958. Tracheal homografts in dogs. *Arch. Otolaryngol.* (Chic.) 65: 577.
- Korppi, H. J. 1970. Tierexperimentelle Untersuchungen mit Reimplantationen kompletter Trachealsegmente. *Arch. Klin. Exp. Ohr Nas Kehlkopfheilk.* 196: 354.
- Tala, P. & Maamies, T. J. 1968. Observations on tracheal reconstruction in experimental animals. *Ann. Chir. Gynaec. Fenn.* 57: 493.
- Toivio, I. T. & Rapo, Seppo E. The role of splenic preservation of transplant rejection. Presented at Suomen Kirurgiyhdistyksen talvikuukausi (Finnish Surgical Society winter meeting), 17 March 1972.
- Toivio, I. T. & Rapo, Seppo E. The role of splenic denervation in homograft acceptance. To be published.
- Toivio, I. T., Rapo, Seppo E., Särälä, U., Tallberg, Th., Laanema, S., Mäkelä, H. & Tapaninen, J. Skin and tracheal homografts following splenic denervation. To be published.

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DISCUSSION

G. Yannoulis: Wollte mir die Frage erlauben: Wo Sie die Optimum-Zeit zwischen der Entnahme der Trachea und ihrer Transplantation bezüglich der Wiederbelebungsfähigkeit festgestellt hätten?

Wir haben nämlich bei unseren Homotransplantationen des Tympano-ossiculären Systems als optimale Zeit, das Ende der zweiten Woche nach der Entnahme für die Transplantation konstatiert. Es möchten wissen, ob vielleicht das gleiche für die Trachea-Homotransplantation, vorkommt.

H. H. Newman: Die interessanten Befunde von Herrn Särälä sind sehr aktuell, denn die plastisch rekonstruktiven Massnahmen an der Trachea werden angesichts der wachsenden Jahre der Tracheektomie immer wichtiger. Sicher besteht kein Zweifel, ein Knorpel ebenso wie Knochen als Homotransplantat ein Antigen ist, wenn auch ein vergleichsweise schwacher. Obion hatte also sicher nicht recht, wenn er glaubte, dass homologer Knorpel unverändert in Wirtsgewebe inkorporiert würde. Wegen der relativ schwachen Immun-Aktivität ist es nun freilich bei derartigen Transplantaten nicht leicht, immunologische Gewebsreaktionen morphologisch sicher abzugrenzen gegenüber normalen entzündlichen Phänomenen. Wir hatten das gleiche Problem, als mein Mitarbeiter Dr. Kastenbauer die immunologische Potenz von homologen Knochen- und Knorpeltransplantaten im Bereich des Mittelohrs untersuchte. Er half sich dadurch, dass er die Akzentuierung der immunologischen Gewebsreaktionen die sogenannte „second-set reaction“ zu Hilfe nahm. Damit wurde die Deutlichkeit der Befunde erheblich besser. Deshalb meine Frage: Haben Sie für die Beurteilung der Antigenität der Trachea-transplantate ebenfalls von der Möglichkeit der „second-set reaction“ Gebrauch gemacht?

T. Petrá: I should like to comment on the views of Toivio in tissue acceptance. It does not take into account the known facts, e.g. that the „killer cells“ the T-lymphocytes, mature by passing through the thymus, or that the human leucocyte antigen is one of the greatest obstacles in tissue transplantation. Nevertheless, my immunological colleague, Dr. Rönkä, told Dr. Toivio to do some spleen denervation for his dogs, and in all of these a classical tissue rejection became apparent. What Dr. Toivio had interpreted as infections were in reality rejection phenomena caused by the lymphocytes.

P. M. Kiviranta: I think it was important to stress that the opinion of Gibson may be observed. Cartilage may also possess immune responses not so important as of skin and muscle.

A second point is the difficulty to have an early idea of the rejection in the control over experimental conditions.

For these reasons we prefer to have biochemical results, such as the dosage of the LDH fractions. In our experiments the only way to have no response in cartilage transplantation is to go through culture of cartilage.

U. Särälä (Reply) to Mr. Yannoulis: Two weeks must be the best interval.

To Mr *Nasomov*. The second-set procedure has been used in our skin transplant studies.

To Mr *Palva* (Rennio): I do not know very much about the experiments in Oulu, but according to what I have heard, the transplanted pieces of skin were badly infected.

To Mr *Klaysen*: I am very thankful for the remarks, which we will consider when continuing our transplantation work. The whole problem of organ transplantation is a very difficult one.

The report given is a preliminary one.

VERIFICATION EXPERIMENTALE SUR L'ANIMAL DE QUELQUES ASPECTS ELECTRO-COCHLEOGRAPHIQUES DE LA PATHOLOGIE HUMAINE

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Abstract Les premiers résultats d'une série d'expérimentations visant à reproduire chez l'animal les différents types de réponses du nerf auditif observés dans la pathologie humaine sont rapportés. 5 cobayes équipés d'une électrode à demeure sur la fenêtre ronde (oreille gauche) ont été soumis à des traumatismes sonores (bruit blanc 110 dB) répétés (6 fois 6 heures). L'évolution des réponses du nerf auditif au clic et aux différents clics filtrés est observée en enregistrant régulièrement avant pendant et après la série de traumatismes. Le microphonique cochléaire est également mesuré au début et à la fin de l'expérimentation. Au moins 5 semaines après le dernier traumatisme des enregistrements de contrôle sont effectués sur les oreilles droites et, après sacrifice du cobaye, les organes de Corti des cochlées droites et gauche sont observés en microscopie à contraste de phase (surface préparations). Chez les 5 cobayes on observe, à la fin de l'expérimentation, des réponses du nerf auditif en 1 point comparables à celles observées chez l'homme dans les cas de réponses dissociées, typiques des surdités professionnelles. Les cochléogrammes reflètent des organes de Corti soit normaux, soit présentant de légères destructions des cellules ciliées externes, principalement au niveau de 2^e et 3^e tours.

Lors de l'enregistrement des réponses du nerf auditif chez l'homme que nous effectuons depuis plusieurs années en tant qu'examen diagnostique périphérique dans la surdité, selon la méthode électro-cochléographique (Aran & Le Bert, 1968 Aran et al. 1971), nous observons très souvent, entre autres, deux types de réponses particuliers qui semblent correspondre à des lésions très caractéristiques de

l'organe de Corti. Ce sont les réponses de types *recrutant* et *dissocié* qui ont été décrits fréquemment et dont deux exemples sont représentés sur les Figs. 1 et 2 (Portmann et al. 1972 Aran 1971).

La comparaison de leurs caractéristiques respectives tant cliniques et audiométriques qu'électro-cochléographiques avec celles des sujets normaux (Fig. 1) suggère immédiatement que l'on est en présence d'une atteinte des structures sensorielles et/ou nerveuses externes généralisée tout le long de la membrane basilaire dans les cas recrutants (Fig. 1) et, dans les cas dissociés, localisée uniquement au premier tour de spire ainsi que l'indiquent l'audiogramme et les réponses du nerf auditif au clic non filtré et au clic filtré aigu (Fig. 2). Les structures moins sensibles (internes) seraient préservées, au moins partiellement, dans les deux cas.

Ce sont ces deux types de réponses que nous avons voulu tout d'abord reproduire chez l'animal afin d'en vérifier le bien fondé des interprétations et d'en approfondir les mécanismes.

Nous rapportons donc ici les premiers résultats d'une recherche à long terme dont l'aboutissement sera, une fois qu'on aura reproduit chez l'animal tous les types de réponses observés chez l'homme et qu'on aura pu y faire correspondre des bilans anatomo-pathologiques précis, un intérêt diagnostique considérable en même temps que très enrichissant.

Chargé de Recherche à l'I.N.S.E.R.M.
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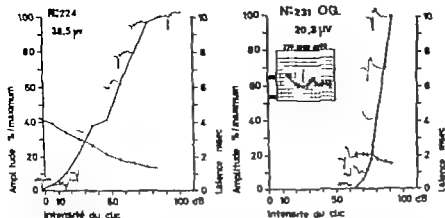


Fig 1 A gauche électro-cochléogramme normal chez un enfant. A droite électro-cochléogramme typique recrutant et audiogramme correspondant. (Stimulations par clic non filtré) (sur cette figure et les sui-

vantes une déflexion vers le bas correspond à la cochlée négative par rapport à l'électrode de référence donnée des traces 10 msec. — db normal (Hearing Level = H. L.)

sur le plan de la connaissance du fonctionnement du récepteur auditif

Désirant reproduire chez le cobaye des réponses recrutantes nous avons d'abord obtenu des réponses dissociées ce qui, compte tenu du fait que celles-ci se situent entre les réponses normales et les réponses recrutantes, reste très logique.

Ce sont les résultats d'une première série expérimentale portant sur 5 cobayes soumis à des traumatismes sonores et dont les réponses sont enregistrées d'une manière tout à fait comparable à l'électro-cochléographie humaine (même matériel, même procédure) que nous décrivons ici.

METHODES

Animaux « chroniques »

Les expérimentations consistent à faire subir à des cobayes normaux des traitements oto-destructifs tels que l'on obtienne des formes de réponses définies à l'avance. Il a semblé que seule l'observation continue des réponses permettait, en contrôlant leur évolution au cours des différents traitements, de doser ces derniers et de les répéter jusqu'à l'obtention des réponses désirées.

Pour cela nous avons travaillé avec des cobayes équipés d'une électrode à demeure sur la fenêtre ronde (oreille gauche) selon une technique mise au point précédemment dans le Laboratoire (Portmann et al., 1966). Les électrodes indifférente et masse étant prises sur les vis servant à maintenir le connecteur cimenté sur le crâne.

Simulations sonores

Une difficulté dans l'utilisation de l'animal éveillé réside dans la manière de délivrer les clics au tympan. Cela est très difficile à réaliser en champ libre car le moindre mouvement de la tête du cobaye modifie à la fois l'intensité et la forme du signal sonore, ainsi que son temps de propagation depuis le haut parleur jusqu'au tympan. Cela est particulièrement incompatible avec la sommation, dans un moyenneur des réponses successives lors de stimulations répétitives.

C'est pourquoi les stimulations sonores sont délivrées directement au tympan par l'intermédiaire d'un tube plastique coudé que l'on introduit dans le conduit auditif externe (Fig. 3) en même temps que l'on branche le pré-amplificateur sur les broches du connecteur fixé sur le crâne et auxquelles sont soudées les électrodes. Le tube est relié à un écouteur

VÉRIFICATION EXPERIMENTALE SUR L'ANIMAL DE QUELQUES ASPECTS ÉLECTRO-COCHLÉOGRAPHIQUES DE LA PATHOLOGIE HUMAINE

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Abstract Les premiers résultats d'une série d'expérimentations visant à reproduire chez l'animal les différents types de réponses du nerf auditif observés dans la pathologie humaine sont rapportés. 5 cobayes équipés d'une électrode à demeure sur la fenêtre ronde (oreille gauche) ont été soumis à des transmissions sonores (bruit blanc 110 dB) répétés (6 fois ■ heures). L'évolution des réponses du nerf auditif au clic et aux différents clics filtrés est observée en enregistrant régulièrement avant pendant et après la série de transmissions. Le microphonique cochléaire est également mesuré au début et à la fin de l'expérimentation. Au moins 5 semaines après le dernier traitement des enregistrements de contrôle sont effectués sur les oreilles droites et, après sacrifice du cobaye les organes de Corti des cochlées droite et gauche sont observés en microscopie à contraste de phase (surface préparations). Chez les 5 cobayes on observe, à la fin de l'expérimentation, des réponses du nerf auditif en 1 point comparables à celles observées chez les cas de réponses dissociées typiques surdités professionnelles. Les cochléogrammes reflètent des organes de Corti soit normaux, soit présentant de légères destructions des cellules ciliées externes, principalement au niveau de 2 et 3 tours.

Lors de l'enregistrement des réponses du nerf auditif chez l'homme que nous effectuons depuis plusieurs années en tant qu'examen diagnostique périphérique dans la surdité selon la méthode électro-cochléographique (Aran & Le Bert, 1968 Aran et al. 1971), nous observons très souvent, entre autres, deux types de réponses particuliers qui semblent correspondre à des lésions très caractéristiques de

l'organe de Corti. Ce sont les réponses types *recrutant* et *dissocié* qui ont été décrits fréquemment et dont deux exemples sont présentés sur les Figs. 1 et 2 (Portmann et 1972 Aran 1971).

La comparaison de leurs caractéristiques respectives tant cliniques et audiométriques qu'électro-cochléographiques avec celles des sujets normaux (Fig. 1) suggère immédiatement que l'on est en présence d'une atteinte des structures sensorielles et/ou nerveuses externes généralisée tout le long de la membrane basilaire dans les cas *recrutants* (Fig. 2) et, dans les cas *dissociés*, localisée uniquement au premier tour de spire, ainsi que l'indiquent l'audiogramme et les réponses du nerf auditif au clic non filtré et au clic filtré aigu (Fig. 2). Les structures moins sensibles (internes) seraient préservées, au moins partiellement, dans les deux cas.

Ce sont ces deux types de réponses que nous avons voulu tout d'abord reproduire chez l'animal afin d'en vérifier le bien fondé des interprétations et d'en approfondir les mécanismes.

Nous rapportons donc ici les premiers résultats d'une recherche à long terme de l'aboutissement sera, une fois qu'on aura produit chez l'animal tous les types de réponses observés chez l'homme et qu'on aura pu faire correspondre des bilans anatomo-pathologiques précis, d'un intérêt diagnostique considérable en même temps que très enrichissant.

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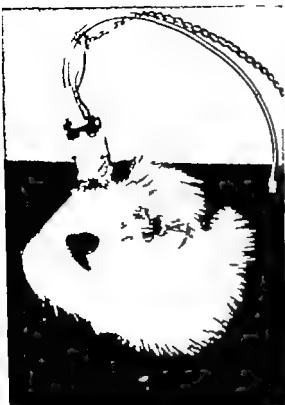


Fig. 3 Cobaye équipé de l'électrode sur la fenêtre ronde (oreille gauche). Branchement des électrodes et du tabe amenant la stimulation sonore de l'écouteur à l'intérieur du conduit auditif externe.

le potentiel d'action global (dans lequel le microphonique cochléaire est éliminé grâce à l'inversion de la stimulation sonore) de 10 en 10 dB depuis le seuil jusqu'à 130 dB. Pour le clic et les clics filtrés 8 000, 4 000 et 2 000 Hz la fenêtre est de 10 msec, avec un délai de 1 msec entre l'ouverture de la fenêtre et l'arrivée du clic au tympan elle est de 20 msec pour le clic filtré 1 000 Hz. Enfin on enregistre également le microphonique cochléaire (alors sans inverser la stimulation) à 110 dB pour le clic et 130 dB pour les clics filtrés avec des fenêtres respectivement de 5, 2, 4, 8 et 16 msec.

Mesures

Comme pour l'électro-cochléographie on mesure sur chaque réponse l'amplitude crête-à-

crête et la latence. Si les latences des pics N1, N2 etc. sont sans ambiguïté dès le seuil sur les réponses normales, ce n'est pas le cas, comme on le verra dans les résultats, dès que les réponses sont déformées. En effet il est à ce moment là plus difficile de reconnaître les différents pics présents dans la réponse. C'est pourquoi, bien que nous mesurions leurs latences respectives, nous ne représentons, sur les courbes entrée-sortie, que le délai entre la stimulation sonore et le point le plus négatif de la trace. Celui-ci correspond alors, suivant les amplitudes relatives des différents pics, à N1 ou à N2 voire N3 etc.

Les valeurs de l'amplitude et de la latence sont mesurées directement sur l'écran de l'oscilloscope et les réponses sont ensuite enregistrées sur papier avec une amplification telle que la dimension du tracé soit toujours sensiblement la même. Cela afin de pouvoir plus facilement comparer les formes des différentes réponses entre elles.

PROTOCOLE EXPÉRIMENTAL

Cinq cobayes normaux, équipés de 1 électrode sur la fenêtre ronde d'une oreille (gauche), ont été soumis à des traumatismes sonores selon le schéma suivant

- 1) Au moins 20 jours après la mise en place de l'électrode les réponses au clic et aux différents clics filtrés sont enregistrées.
- 2) Les cobayes sont alors exposés pendant 6 heures à un bruit blanc de 110 dB SPL (TS1 premier Traumatisme Sonore).
- 3) Pendant les 7 jours suivants, l'évolution des réponses (récupération) est observée, en enregistrant les réponses au clic.
- 4) Les cobayes sont ensuite exposés de nouveau 5 jours de suite au même bruit blanc (6 heures, 110 dB) (TS2 à TS6). Ils sont testés chaque jour avant l'exposition au bruit.
- 5) Les réponses au clic sont ensuite enregistrées régulièrement jusqu'à ce qu'elles cessent d'évoluer (environ pendant un mois après le dernier traumatisme)
- 6) La veille du jour où chaque

sacré, on enregistre les réponses de l'oreille équipée (gauche) pour l'ensemble des stimulations (clic et clics filtrés).

7) Le lendemain l'oreille droite est testée de la même manière (électrode sur la fenêtre ronde) sous anesthésie générale.

8) L'animal est ensuite sacrifié et les deux cochlées sont fixées après décapitation (glutaraldéhyde, tampon, acide osmique). Les cochléogrammes pour les oreilles droite et gauche sont alors réalisés en microscope à contraste de phase (*surface preparations* Engström et al. 1964) sur des fragments des différents tours prélevés dans la moitié la plus accessible de la cochlée. L'autre moitié de la cochlée est incluse dans l'Epon pour observation ultérieure en microscope électronique.

Nous ne rapportons ici que les résultats des cochléogrammes en contraste de phase, les observations en microscope électronique étant en cours.

RÉSULTATS

Tout d'abord mentionnons que 3 cobayes normaux témoins, équipés de la même manière d'une électrode à demeure sur la fenêtre ronde ont été simultanément observés 2 pen-

des durées de 109 et 122 jours respectivement jusqu'au sacrifice pour contrôle histologique. Les réponses sont restées, sur le plan des seuils, des formes et des courbes, identiques. Les cochléogrammes ont révélé des organes de Corti normaux à l'exception du 4^e tour où quelques cellules ciliées externes étaient absentes, cela de la même manière pour les cochlées gauches (équipées en permanence de l'électrode) et droites (non concernées par l'approche chirurgicale de l'oreille gauche ni par les stimulations sonores lors des différents enregistrements). Le 3^e cobaye non encore sacrifié, présente, après plus de 15 mois, des réponses normales comme aux premiers jours.

Soulignons aussi que les enregistrements effectués le jour du sacrifice sur l'oreille non équipée (droite) ont été chaque fois très com-

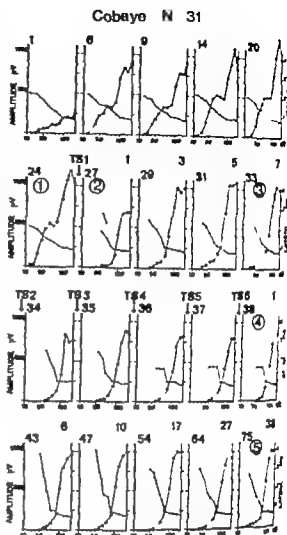


Fig. 4. Evolution des réponses au clic (courbes entrée-sortie, amplitude-latence) chez le cobaye N° 31. Les chiffres en haut à gauche de chaque graphique expriment le nombre de jours après la mise en place de l'électrode sur la fenêtre ronde. Les chiffres en haut à droite expriment le nombre de jours après le dernier traumatisme sonore précédent. T81 T82 etc. traumatismes sonores successifs. Les chiffres encadrés signalent les 5 étapes importantes dans l'expérimentation: 1 la veille du premier traumatisme (T81) 2 le lendemain de T81 3 7 jours après T81 4 le lendemain de T86 5 la veille du sacrifice, soit moins 5 semaines après T86.

parables à ceux réalisés la veille pour l'oreille gauche chez l'animal éveillé. On remarque seulement une légère élévation des seuils par rapport à ceux de l'oreille gauche (due certainement à l'approche chirurgicale de l'oreille

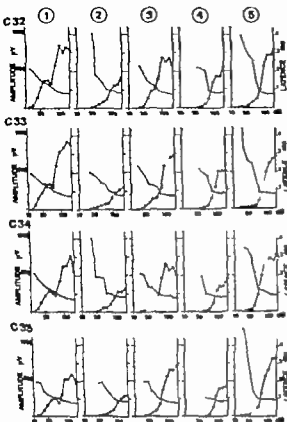


Fig 5 Courbes entrée-sortie (i.e. non filtré) pour les autres cobayes, aux 5 étapes définies dans la figure 4

juste avant les enregistrements) et une légère, mais constante, augmentation de la latence des réponses (dépendant de la profondeur de l'anesthésie, comme on a pu le vérifier en observant les réponses à gauche sur animal anesthésié). Nous ne rapportons donc pas dans le détail les résultats des enregistrements effectués sur les oreilles droites, destinés essentiellement à contrôler la validité des observations faites sur les oreilles gauches. Par contre ces dernières sont présentées presque exclusivement car outre leur intérêt dans le cadre de notre recherche elles permettent de montrer les possibilités de cette technique expérimentale qui devrait s'avérer très pratique pour l'étude de nombreux phénomènes auditifs volutifs.

Evolution des Réponses au Clic

Les réponses normales avant le premier traumatisme

Bien que les amplitudes des réponses puissent être d'un cobaye à l'autre, assez différentes (certainement en fonction de la qualité des électrodes), les formes des réponses et des courbes entrée-sortie, amplitude-latence sont absolument comparables. Du seuil (autour de 20 dB peSPL) au maximum (130 dB peSPL) la latence varie progressivement de 2 msec à 0,8 msec; l'amplitude augmente, avec l'intensité du clic, en deux étapes avec le plateau intermédiaire autour de 80 dB (Fig. 4 première rangée, Fig. 5-1) les différents pics N1 N2 etc. sont nettement visibles dans les formes des réponses dès le seuil (Fig. 6-1).

Effets du premier traumatisme sonore (TSI)

Le lendemain du premier traumatisme sonore on observe les modifications suivantes dans les réponses au clic (Fig. 4 5-2 et 6-2)

Le seuil (autour de 50 dB peSPL) s'est élevé en moyenne de 30 dB.

La latence au voisinage du seuil, de l'ordre de 2 msec, est de ce fait plus élevée par rapport à sa valeur aux mêmes intensités avant le traumatisme il en est ainsi jusqu'à 30 à 40 dB au dessus du seuil, au delà la latence est la même.

L'amplitude augmente lentement jusqu'à 80 dB puis plus rapidement au-delà, on n'observe plus le plateau autour de 80 dB (forme « exponentielle » pour la courbe amplitude) au maximum (130 dB) l'amplitude est réduite d'environ 50%.

Les réponses sont considérablement élargies près du seuil et les pics N1 et N2 ne sont plus décelables.

Aux fortes intensités les pics négatifs sont nets mais précédés d'une déflexion positive importante. Celle-ci s'observe également avant le traumatisme elle représente vraisemblablement le potentiel de sommation (positif car on enregistre au niveau de la rampe tympanique). Cependant le pic positif et la réponse

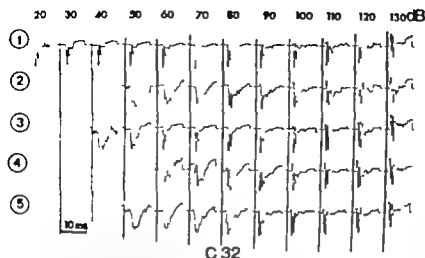


Fig 6 Formes des réponses au clic chez le cobaye N° 3 lors des 5 étapes définies dans la figure 4 sauf pour la dernière rangée qui correspond aux enregistrements effectués 17 jours au lieu de 39 après TS6. (Les tracés ont sensiblement la même dimension afin

de comparer plus facilement entre elles les formes des différentes réponses. Pour les amplitudes, se reporter aux courbes correspondantes sur la figure 5-cobaye 32, sauf pour la dernière rangée 5.)

nervieuse ont tous les deux diminué après le traumatisme mais dans des proportions différentes.

Récupération après le premier traumatisme

Après 7 jours de récupération (Fig. 4 5-3 et 6-3)

Le seuil n'est plus qu'à 10 dB environ du seuil normal.

Les courbes entrée-sortie amplitude-latence ne sont pas cependant redevenues normales. Elles sont à faible amplitude et latence longue en dessous de 80 dB. Si trois cobayes présentent toujours des courbes amplitude « exponentielles » chez les deux autres (32 et 33) on observe la réapparition du plateau autour de 80 dB mais son amplitude est plus faible.

Aux fortes intensités l'amplitude, la latence et la forme des réponses sont pratiquement les mêmes qu'avant le traumatisme.

Près du seuil les formes des réponses sont cependant toujours larges (Fig. 6-3).

Effets des 5 traumatismes suivants (TS2 à TS6)

Le premier jour après les 5 traumatismes sonores successifs (Fig. 4 5-4 et 6-4) on note que

Le seuil n'est que 10 dB environ au-dessus de ce qu'il était le lendemain du premier traumatisme (60 à 70 dB)

La latence est toujours autour de 2 msec seuil (sauf pour le cobaye 35 où elle d'emblée de 0,8 msec). Elle passe brusquement de 2 à 0,8 msec 30 dB au-dessus du seuil, reste sensiblement constante jusqu'à 130 dB.

L'amplitude maximum est légèrement plus grande qu'après le premier traumatisme.

Les formes sont toujours larges près du seuil et paraissent normales au maximum. On remarque, aux niveaux intermédiaires, l'apparition progressive de N1 (Fig. 6-4 à 70, 80, 90 dB)

Evolution après le dernier traumatisme

Les réponses sont très caractéristiques et se stabilisent pour les 5 cobayes (Fig. 4 5-5, 6-5). Elles sont pratiquement stabilisées vers 20^e jour après le dernier traumatisme (Fig. 4 On note que

Le seuil se stabilise autour de 40 dB.

La latence est nettement plus longue près du seuil (entre 3 et 4 msec) et elle décroît rapidement jusqu'à 80 dB où elle reste constante autour de 0,8 msec, cela dès les premiers jours après TS6 (Fig. 4)

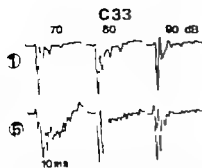


Fig 7 Détail des réponses au clic chez le cobaye N° 33 au début (D) et à la fin (E) de l'expérimentation. Noter l'apparition progressive de N1 de 70 à 80 et 90 dB.

L'amplitude est relativement très faible en dessous de 80 dB puis elle augmente très rapidement et est, à 130 dB, pratiquement la même qu'avant les traumatismes.

Les formes sont toujours larges aux faibles intensités et apparemment normales au maximum. On observe encore très nettement l'apparition progressive du pic N1 précédant la réponse tardive large, lorsqu'on augmente l'intensité du clic. (Fig. 6-5 et 7).

Réponses au clic et aux différents clics filtrés avant et après les traumatismes

Les réponses aux différents clics filtrés n'ont été observées qu'au début et à la fin de l'expérimentation (Fig. 8 et Tableau I).

Les réponses aux clics filtrés basse fréquence 1 000 et 2 000 Hz ne sont pratiquement pas sur le plan des seuils, de l'amplitude de la latence.

Par contre on note une très nette élévation des seuils pour les clics filtrés aigus 8 000 et 000 Hz.

La latence au seuil pour les clics filtrés aigus, soit à 8 000 Hz, soit à 4 000 Hz, courte ≈ 1 msec. Ainsi, pour le cobaye 31 (Fig. 8) le seuil pour le clic filtré 8 000 Hz est passé 40 à 90 dB la latence de 2 à 1 msec et la 1re partie de la courbe a disparue alors l'amplitude maximum à 130 dB est identique. Pour le clic filtré 4 000 Hz le seuil est modifié et on distingue dans les courbes

amplitude-latence les deux segments distincts de part et d'autre de 90 dB. On note encore de légères modifications pour le clic filtré 2 000 Hz et pratiquement aucune pour le 1 000 Hz.

La diminution du microphonique cochléaire sur les réponses au clic et aux clics filtrés, exprimée en dB sur le tableau 1 est faible. Elle est en général plus prononcée pour les clics filtrés 4 000 et 2 000 Hz.

Bilan histologique

La proportion des cellules sensorielles détruites par rapport à l'ensemble des cellules ciliées est extrêmement faible. A titre d'exemple les figures 9 et 10 montrent les cochléogrammes des deux oreilles du cobaye le moins atteint sur le plan Ià (N 31) et de celui qui présente le plus de destructions (N 33). Le tableau 1 exprime le nombre de cellules ciliées externes détruites, en % du nombre total de cellules externes, pour les différents tours, chez les 5 cobayes. On remarque qu'en moyenne les destructions se trouvent principalement au niveau du 2^e tour. Comme on pouvait s'y attendre, les cellules ciliées internes sont intactes

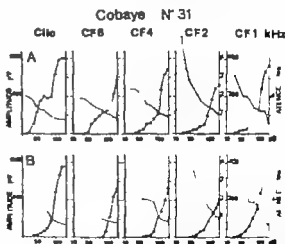


Fig 8 Comparaison des courbes entrées-sorties pour le clic et les clics filtrés au début (A) et à la fin (B) de l'expérimentation chez le cobaye N° 31. Remarquer la similitude des courbes pour le clic et le clic filtré 8 000 Hz, en (A) avec celles d'un sujet humain normal (Fig. 1 gauche, pour le clic) et, en (B), avec celles d'un sujet présentant des réponses dissociées (Fig. 2 pour le clic et le clic filtré 8 000).

Tableau I. Comparaison, pour les 5 cobayes des modifications du microphonique cochléaire (ΔMC perte en dB), et des seuils du potentiel d'action ($\Delta S.P.A$ élévation du seuil en dB), pour le clic et les clics filtrés, avec les destructions de cellules ciliées externes (ΔCCE en % pour l'ensemble des tours (total) et aux différents tours (T1 T2 T3 et T4)

ΔMC dB	Clic		8 kHz		4 kHz		2 kHz		1 kHz						
$\Delta S.P.A$ dB	Clic		8 kHz		4 kHz		2 kHz		1 kHz						
ΔCCE (%)	Total		T1		T2		T3		T4						
31	12,6	20	2,1	6,3	30	3,1	10,4	20	1,2	11,1	20	2,6	4,4	0	1
32	3,3	15	6,3	2,3	20	1,4	0,8	30	10,3	2,6	30	15,2	1,3	0	2
33	14	10	12,3	8,2	30	3,2	9,1	30	39,7	9,1	30	10	6	10	5
34	14	20	3,4	6,2	40	1,8	3,5	30	14,1	11,5	30	14,1	4,3	0	
35	9,5	10	9	9,1	45	6,5	11,1	40	8,7	10,1	10	10,9	4,6	5	9

(une seule cellule détruite sur l'ensemble des fragments examinés dans les 10 cochlées des 5 cobayes).

DISCUSSION

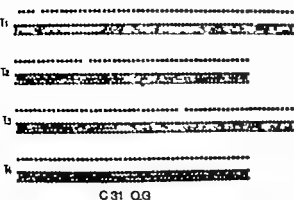
Si toutes les données expérimentales ont été décrites avec le plus de détails possible seules les caractéristiques les plus importantes seront discutées ici, en fonction de l'objectif particulier de ce travail. En effet certaines particularités ne pourront être efficacement discutées que lorsqu'on disposera de données en plus grand nombre et plus diversifiées à l'occasion traitements oto-destructifs variés, en particulier plus sélectifs que le bruit blanc à large bande.

Tout d'abord soulignons que nos observations étaient principalement centrées sur les aspects et l'évolution des formes des réponses et des courbes entrée-sortie, ainsi que sur les bilans anatomiques. La détermination des seuils du potentiel d'action et la mesure du microphonique cochléaire n'ont pas été effectuées avec un souci de grande précision. Etant donné d'autre part le caractère relativement modéré de l'agression sonore et les faibles variations des seuils du potentiel d'action et de l'amplitude du microphonique, les résultats, en ce qui concerne ces deux grandeurs, sont moins significatifs qu'ils ne le seraient lors de traitements otodestructifs plus importants. Sur ces

deux plans là on peut conclure cependant, en observant le tableau I que les modifications sont notables pour les clics filtrés 8 000 et 4 000 Hz, légères pour le 2 000 Hz et très faibles pour le 1 000. Cela impliquerait une atteinte préférentielle des premiers tours de la cochlée.

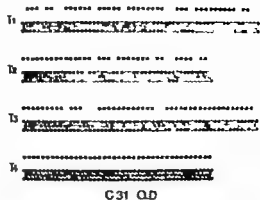
Sur le plan des formes des réponses et des courbes entrée-sortie les modifications sont beaucoup plus nettes et remarquablement comparables pour les 5 cobayes. On retrouve en outre toutes les caractéristiques des réponses « dissociées » décrites chez l'homme (Fig. 7). Si on admet que les traumatismes sonores utilisés (bruit blanc à 110 dB) et leur répétition sont comparables à ce qui peut exister dans certains lieux de travail on peut admettre aussi que les mécanismes d'induction de ces modifications sont les mêmes dans notre expérimentation que chez l'homme présentant les réponses « dissociées » qui sont typiques ainsi que l'audiogramme, des surdités d'origine professionnelle.

Les lésions des cellules ciliées externes telles qu'on peut les observer au microscope à contraste de phase apparaissent très modérées. Bien que réparties sur l'ensemble des tours elles sont prédominantes au niveau des 1^{er} et 3^{es} tours, ce qui est en parfait accord avec le contenu spectral du bruit blanc et les propriétés mécaniques de la membrane basilaire. Le fait que ces lésions ne soient pas



C 31 O.G

Fig. 9 Cochléogrammes des cochlées droite et gauche cobaye N° 31. Des 5 cobayes, celui-ci présente le



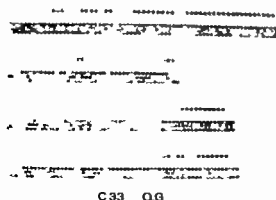
C 31 O.D

moins de destructions cellulaires. e, cellule ciliée détruite.

localisées indiquerait en outre que les réponses « dissociées » apparaissent dès qu'il y a des lésions des cellules externes, quelle que soit leur localisation. Cependant le fait que les réponses soient altérées de la même manière lorsque l'organe de Corti paraît normal (cobaye 1 Fig. 9) que lorsqu'il est effectivement lésé (cobaye 33 Fig. 10) suggère que ou bien des cellules ciliées apparemment normales (d'autant plus que le temps de récupération après le dernier traumatisme excède 1 mois) peuvent ne pas être fonctionnelles, ou bien que les lésions motrices se situent à un autre niveau (au niveau des terminaisons nerveuses?), inaccessible au microscope à contraste de phase. In (1971) a décrit les différents niveaux où on pouvait effectivement observer

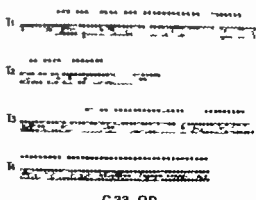
au microscope électronique, des altérations immédiates après le traumatisme sonore. Ce pendant pour des intensités de 110 dB pendant 2 heures, il ne note aucune modification. Il est certain que, dans notre expérimentation, la durée du premier traumatisme (6 heures), leur répétition et le délai respecté avant le sacrifice ont induit des modifications définitives.

Enfin il est intéressant de noter que, sur le plan des seuils comportementaux chez l'animal, on s'accorde à trouver pas ou peu de modifications permanentes des seuils après traumatisme sonore modéré, avec cependant des altérations certaines des cellules ciliées externes (Ward & Duvall, 1971; Miller et al., 1971). Par contre nous trouvons, pour des lésions minimales, voire nulles, de l'organe de Corti, une légère éléva-



C 33 O.G

Fig. 10 Cochléogrammes des oreilles droite et gauche cobaye N° 33. C'est celui qui présente, parmi les



C 33 O.D.

5 cobayes, le plus de destructions cellulaires. e, cellule ciliée détruite.

tion des seuils électrophysiologiques mais sur tout, et cela nous paraît le fait le plus important qui dépasse le seul problème de l'audition liminaire, une modification nette et significative des formes des réponses et des courbes entrée-sortie. Celles-ci impliquent une modification dans la synchronisation des fibres nerveuses et en conséquence dans la sensation auditive supra-liminaire.

SUMMARY

The first results are reported of a series of experiments designed to reproduce in the animal the different patterns of VIII nerve responses observed in human pathology. Five guinea pigs with an electrode permanently implanted on the round window (left ear) are exposed to repeated acoustic trauma (6 times 110 dB white noise for 6 hours). VIII nerve responses to click and filtered clicks are followed before, during and after the acoustic traumas. Cochlear microphonic is also measured at the beginning and the end of the experiment. Five weeks (minimum) after the last acoustic trauma, control recordings are performed on the right ear before sacrifice and the right and left cochleas are observed by phase contrast microscopy (surface preparations). At the end of the experiment the VIII nerve responses are similar to the "disassociated" type of response observed in man in cases of occupational deafness. The organs of Corti range from normal to mildly injured at the level of the outer hair cells, mainly at the 2nd and 3rd turns.

ZUSAMMENFASSUNG

Die Resultate von Untersuchungen um beim Tier globale Wirkungen des Gehörnervs wieder zu erzeugen, vergleichbar den elektro-cochleographischen Wirkungen „disassociated“ beim Menschen, liegen vor Meerschweinchen, welche mit einer Dauer Elektrode auf dem runden Fenster ausgestattet sind, wurden gemässigten obstruktiven Behandlungen unterworfen (Klangverletzung). Die Antwort auf Klick

(charakteristische Eingang-Ausgang- Weite-Latenz Formen) wird während und nach der Behandlung beobachtet. Das histologische Bild in der kontrast mikroskopischen Untersuchung wird besprochen in Lässch der elektro-physiologischen Angaben.

REFERENCES

- Aran, J.-M. 1971 Clinical measures of VIII nerve function. Int. Symp. on Oto-physics (4: Arbor May 1971), in *Advances in O.R.L.* (in press).
- Aran, J.-M. & Le Bert, G. 1968 Les réponses nerveuses cochléaires chez l'homme: image du fonctionnement de l'oreille et nouveau test d'audiométrie objective. *Rev. Laryng. (Bord.)* 89: 361.
- Aran, J.-M., Pelletier, J., Lepoir, J., Portmann, M., Darrouzet, J. 1971 Aspects théoriques et pratiques des enregistrements électro-cochléographiques selon la méthode établie à Bordeaux. *Rev. Laryng. (Bord.)* 92: 601.
- Engström, H., Ades, H. W. & Hawkins, J. E. 1964 Cytoarchitecture of the organ of Corti. *Acta Otolaryng. (Stockh.)*, Suppl. 188: 192.
- Miller, J. D., Rothenberg, S. J. & Eldredge, D. A. 1971 Preliminary observations on the effects of exposure to noise for seven days on the hearing and inner ear of the chinchilla. *J. Acoust. Soc. Amer.* 50: 1199.
- Portmann, M., Aran, J.-M. & Le Bert, G. 1966 Mise en place définitive d'une électrode sur la fenêtre ronde du cobaye. *CR. Soc. Biol. (Par.)* 160: 351.
- Portmann, M., Aran, J.-M. & Lagouge, P. 1971 Testing for recruitment by electrocochleography. Preliminary results. *Ann. Otol.* (in press).
- Spoendlin, H. 1971 Primary structural changes in the organ of Corti after acoustic overstimulation. *Acta Otolaryng. (Stockh.)* 71: 166.
- Ward, W. D. & Duvall, A. J. 1971 Behavioral and ultrastructural correlates of acoustic trauma. *Ann. Otol.* 80: 881.

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ANALYSIS OF BONE REMODELLING IN THE OTOSCLEROTIC STAPES BY MEANS OF TETRACYCLINE LABELLING

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Abstract The distribution and the amount of bone tissues formed in the otosclerotic focus during 6 days were studied in 16 otosclerotic and 3 normal stapedes, by means of tetracycline labelling. The relative amount fluorescent new-formed bone was expressed as a ratio $NB/(NB+PB)$ of new bone (NB) on the pre-existing bone (PB). In the more active foci a layer of new-formed bone was noticed around nearly all the vascular cavities. In the less active and quiescent foci the new-formed bone was only noticed on the edge of some osteons. In the normal stapedes a trace of new-formed bone was observed.

The present investigation was undertaken to study the amount and distribution of the newly formed bone in the otosclerotic focus, using tetracyclines as tracers. The observation by Huet et al. (1958) that tetracyclines become incorporated into bone matrix during its mineralization, enables us to distinguish the newly formed bone from the pre-existing unlabelled bone. In the ground sections the newly formed bone is easily detected under ultraviolet (UV) light owing to the yellow fluorescence of the tetracyclines. Part of this study was reported at the 10th Meeting of the Inner Ear Biology Group, London in London September 1972.

MATERIAL AND METHOD

Twelve stapedes removed during stapes surgery for relief of a conductive deafness pre-oper-

atively diagnosed as clinical otosclerosis were examined. Three normal stapedes removed from patients operated on by labyrinthectomy for Ménière's disease served as controls.

To all the subjects 900 mg demethylchlor tetracycline (Ledermycin) were orally administered for 5 successive days. With this dosage the tetracycline blood level was kept sufficiently high to allow an appreciable fixation to newly formed bone tissue during 6 days. The stapedes were removed 10 days after the last tetracycline administration. During this period of time the tetracycline completely disappears from the connective tissue while it remains in the newly formed bone tissue.

The stapedes were fixed in alcohol, dehydrated and embedded in methacrylates. After polymerisation the specimens were ground by hand to a thickness of 40-50 μ . The sections were photographed under UV light on colour reversal film (Ektachrome High Speed) using the Zeiss Ultraphot II microscope equipped with one or two energizing filters BG 12, and one barrier filter OG 50.

On the colour diapositive of each section, the fluorescent new bone (NB) formed during the administration of the tracer, the pre-existing unlabelled bone (PB), and the spaces not occupied by bone tissue were determined at the microscope by means of a Swift automatic point counter with a horizontal spacing of 1/20 mm and a vertical spacing of 1/6 mm.

This research was supported partly by a grant from Consiglio Nazionale delle Ricerche and partly by a grant from Ministero della Sanità.

The values obtained were expressed as percentage ratio of NB/(NB+PB). This ratio expressed the volume of bone tissue which was being added per unit time (amount of bone formation). The values were reduced by 1/6 in order to obtain the amount of bone formation per day.

The sections were microradiographed using a Machlett type AEG 50T X ray tube (7.8 kV 18 mA filter 1 mm Be) to check the mineralization degree of the labelled areas.

RESULTS

Normal stapedes

No fluorescent newly formed bone was observed in any of the normal stapedes (Fig. 1 A). Only a faint green-yellow autofluorescence was present in the articular cartilage for the lenticular process of the incus, the cartilaginous layer of the footplate, the stapedius tendon and in some mucosal remnants (Fig. 1 A). This faint green yellow autofluorescence could easily be distinguished from the yellow fluorescence due to actual binding of tetracycline to bone tissue (Fig. 1). Since the autofluorescence was observed in stapedes from cadavers without a clinical history of tetracycline treatment it could be inferred that it was not related to the administration of the drug.

We concluded that in the 3 normal stapedes the amount of bone formation per day was 0.0 (Table I).

Otosclerotic stapedes

In the normal areas of the sections of otosclerotic stapedes we never observed fluores

Table I "Normal" stapedes

No.	Side	Age (years)	Sex	NB (NB + PB) per day
417	Left	57	♂	0.0
418	Left	18	♀	0.0
290	Right	33	♂	0.0

cent newly formed bone. A faint green-yellow autofluorescence was found in the same locations where it was demonstrated in normal stapedes (Fig. 1 B).

The otosclerotic foci were sub-divided into three groups according to their "activity" as indicated from the histological picture. This evaluation was based upon the dimensions of the marrow spaces, the number, shape and dimension of the cavities and the characteristics of the bone matrix. A single stapes was inserted in more than one group when the foci with a different degree of activity were present.

The more active foci showed a great amount of irregular vascular spaces, enclosed in a network of thin trabeculae consisting of "woven bone". The highest percentage of tetracycline labelling occurred in these foci (Figs. 2, 3). In one exceptional case all the surfaces were fluorescent and a diffuse fluorescence was also present inside the bone tissue. This condition prevented a precise determination of the amount of bone formation. In 4 cases most of the surfaces were labelled with tetracycline (Figs. 2 B, C, and 3) the percentage of labelling varied from 13 to 30% which means that the amount of bone formation per day ranged from 2.0 to 5.0% (Table II).

Table II "Active" otosclerotic foci

No.	Side	Age (years)	Sex	Hearing loss (300, 1000, 2000 Hz)	Evolution (years)	Tinnitus	NB (NB + PB) per day
414	Left	76	♂	45-45-50	4	Present	Very high, not measurable
422	Right	39	♀	65-70-70	15	Present	5.0
431	Right	15	♀	65-65-90	3	Present	2.8
432	Left	11	♀	70-65-60	11	Present	2.6
167	Right	17	♀	55-55-60	3	Present	2.4
429	Left	65	♀	90-75-80	1	Absent	2.0



Fig. 1 (A) Micrograph under UV light of a normal stapes from an 18-year-old patient. (B) Otosclerotic stapes from a 25-year-old patient, photographed in the same conditions (22) and (C) at higher magnification (50). In the normal stapes a faint green-yellow autofluorescence is visible in the cartilage and connective tissue. No trace of fluorescent newly

formed bone can be seen. In the otosclerotic stapes a bright yellow fluorescence marks the newly formed bone. Only in colour pictures (Fig. 1) can the yellow fluorescence due to tetracycline be distinguished from the autofluorescence: in black and white pictures (Figs. 2, 3, 4 and 5) this distinction is not possible.

Table III. "Less active" otosclerotic foci

No.	Side	Age (years)	Sex	Hearing loss (500, 1000, 2000 Hz)	Evolution (years)	Tinnitus	NB (NB + PB) * per day
423	Right	38	♀	85-65-55	13	Present	2.2
422	Right	39	♀	65-70-70	15	Present	2.2
411	Right	46	♂	70-65-80	4	Present	2.2
41	Left	47	♂	70-75-95	17	Present	1.4
434	Right	35	♀	70-70-45			1.4
33	Right	57	♀	45-55-55	7	Present	1.3
427	Left	23	♀	30-40-50	5	Present	1.0
405	Right	25	♂	60-60-60	1	Absent	1.0

The less active foci showed less numerous and smaller vascular spaces (Figs. 1 B C, and 4). In some areas there was a tendency to a lamellar arrangement. A layer of labelled bone lined the wall of only some vascular spaces (Fig. 4). The thickness of this layer was not uniform. Therefore it was possible to distinguish in these foci the active surfaces undergoing calcification from the inactive surfaces of bone deposited prior to administration of tetracycline (Fig. 4 C). In the less active foci the daily amount of bone formation ranged from 1.0 to 2.2% (Table III).

In the quiescent foci the number and dimensions of the vascular spaces were markedly smaller. Haversian systems could frequently be observed, but only a few were labelled with tetracycline (Fig. 5). The amount of bone formation per day was 0.2 to 0.4% (Table IV).

In most of the otosclerotic foci studied, the newly formed bone was observed inside the focus, seldom on the peripheral contour (Fig. 2 C arrows).

DISCUSSION

Several investigators have attempted to study otosclerotic bone deposition by using tetra-

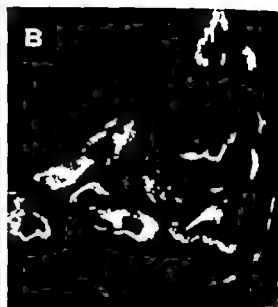
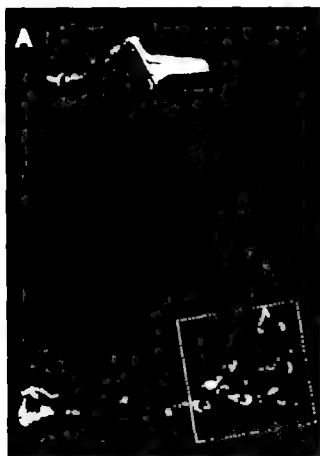
cycline labelling. In 2 patients who had received tetracyclines, 1 year and, respectively 2 years prior to the operation, Frost (1962) observed the remodelling pattern of otosclerotic bone. The most noticeable findings, obtained by Alberti & Tarkannen (1963) using the same technique, were the presence of small amounts of fluorescent material both in the most active and in the most inactive foci. No fluorescence was evident in one normal foot plate.

Clarke's (1965) results on tetracycline-induced fluorescence seem to indicate that there are differences depending on the stage of the disease but his micrographs are questionable. Very little information was obtained with tetracycline labelling by Röckert et al. (1965) and by Crifo (1970). García Ibañez (1966) observed traces of bone deposition in 27 out of 81 fragments of otosclerotic stapedes.

Our results confirm that in the normal stapedes there is no trace of newly formed bone. This observation is in agreement with the findings by Roberto & Morra (1972) who studied the post-fetal bone formation in the auditory ossicles of the dog by means of alizarine labelling. In the dog stapes, only deposition of primary bone tissue was observed and

Table IV. "Quiescent" otosclerotic foci

No.	Side	Age (years)	Sex	Hearing loss (500, 1000, 2000 Hz)	Evolution (years)	Tinnitus	NB (NB + PB) * per day
413	Left	47	♂	80-85-70	12	Present	0.4
433	Right	39	♀	65-60-60	9	Present	0.3
405	Right	25	♂	60-60-60	1	Present	0.3
41	Left	33	♀	70-65-60	1	Absent	0.2



it was completed within the third months of post-natal life. This means that bone remodelling does not take place in the stapes as in other parts of the skeleton.

All the otosclerotic foci showed a more or less active bone deposition which was massive in only one very active case where a high percentage of labelled bone was observed. Apart from this the bone formation rate per day ranged from 2.0 to 5.0% in the active foci, from 1.0 to 2.2% in the less active foci and from 0.2 to 0.4% in the inactive foci.

There is no doubt that the method employed by us only permits an approximate measurement of the amount of tetracycline-labelled bone. Our data should be interpreted with caution also because they represent a 6-day exploration of a pathological process which evolves through many years.

The amount of bone formation per day in active otosclerosis seems high if we consider the long evolution of the disease and if we compare the figures obtained in otosclerosis with the data of the literature on normal and pathological bone. Frost et al. (1960) and Frost (1961) obtained values of 0.01 to 0.02% in normal adult cortical bone and of 0.3 to 0.5% in ribs from young children. Values for the rate of bone formation in the Paget areas ranged from 0.1 to 0.6% per day while in the uninvolved areas they were from 0.04 to 0.1% (Sissons & Lee, 1964). An increased bone formation rate was observed in hyperparathyroidism by v der Stuyt Veer et al. (1964) and in 2 children suffering from osteogenesis imperfecta (from 0.30 to 0.42% per day) by Lee (1965).

Absence of labelled perosteal bone was noted in most of the otosclerotic foci examined by us. This means that the otosclerotic foci undergo an internal remodelling also when

their overall size does not increase. In one case we observed a perosteal deposition in the area adjacent to a blood vessel (Fig. 2C) and this can give an idea of the way the focus increased its size.

The distribution of labelled bone was not uniform inside the same focus, i.e., some areas are more active others less active or quiescent.

It was not possible to find a correlation between the amount of bone deposition and clinical data (see Tables II, III, IV). With regard to the age it can be noticed that the 2 younger patients (15 and 17 years old) belong to the group of high bone deposition rate the same group, however, comprises a patient 65 years old. Relatively young patients showed a low deposition rate. Regarding the duration of the otosclerotic process, 4 recent cases and 2 with a long clinical story belong to the group with very active foci. 3 other cases with a long evolution showed quiescent foci. No relationship could be established between bone deposition rate and hearing loss or tinnitus.

In Spain, stapedectomies were carried out by L. Garcia Ibañez and in Italy by E. Bocca and S. Iurato.

ACKNOWLEDGMENT

We would like to express our thanks to Professor R. Amprino and Professor G. Marotti for their cooperation.

RÉSUMÉ

Par la technique de marquage avec la tétracycline on a analysé la distribution et la quantité du tissu osseux se déposant au cours de 6 jours de traitement dans 16 étiéres otosclérotiques et 3 étiéres normales. La quantité relative de tissu osseux formé au cours du traitement par la fluorochrome a été indiquée par le rapport pour cent entre l'os nouveau (ON) et l'os pré-existant (OP), soit ON/(ON + OP). Dans les coupes des foyers les plus actifs une bande de tissu osseux récemment formé tapisse la paroi de presque toutes

Fig. 2. Otosclerotic stapes from a 65-year-old patient. The distribution and the amount of the newly formed bone labelled with tetracycline is detected under UV light. Almost all the vascular spaces are lined with fluorescent bone. The autofluorescence of the cartilage and of the stapedius tendon cannot be dis-

tinguished from the true fluorescence in a black and white picture. A (×22); inset is shown at higher magnification in B (×65) and C (×130). The picture in (B) was taken with two, and that in (C) with one energizing filter to show that tetracycline labels the bone and not the connective tissue.

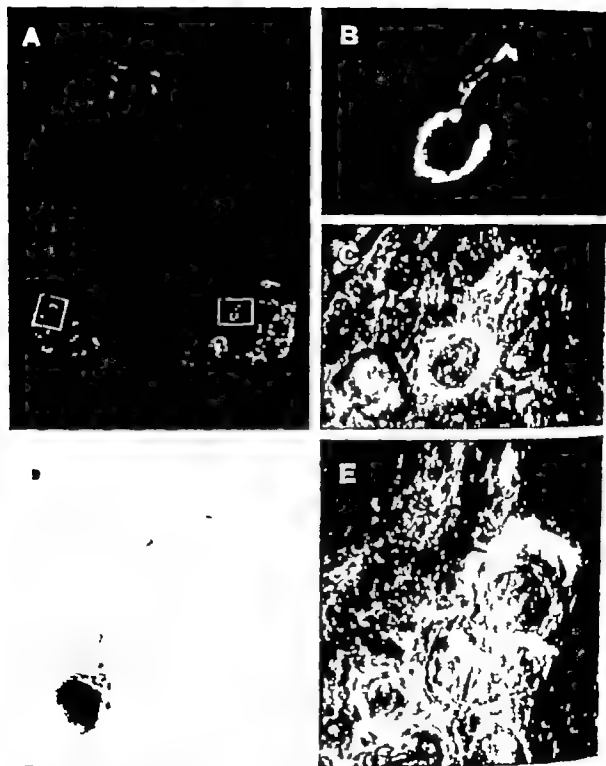


Fig. 3 Otosclerosis, stapes from a 15-year-old patient. "Active" otosclerosis like that shown in Fig. 1. (A, 22, B and C, 19 \times B and E, 43 \times .) The pictures

(C) and (E) were taken with one eretipung (C) to show the structure of the labelled bone and the surrounding area. Macrographs obtained under UV light

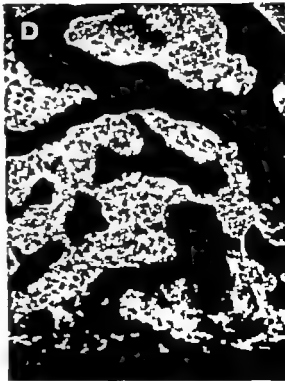
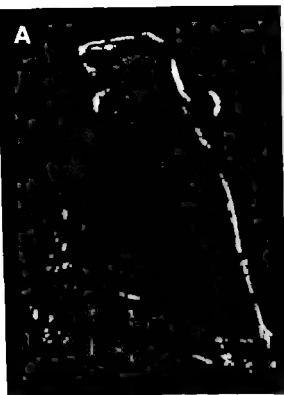


Fig 4 Otosclerotic stapes from a 25 year-old patient. The disease is "less active" than in the cases shown in Figs 2 and 3. (A) The otosclerotic focus involves the whole footplate, but only a small part is labelled with tetracycline (inset). This is shown at $\times 190$ in (B) (2

energizing fibers); (C) (1 energizing fiber); and (D) (microradiograph). In (C) it is possible to distinguish this active surface undergoing calcification from the inactive surfaces and (D) shows the low mineral content of the newly formed bone.

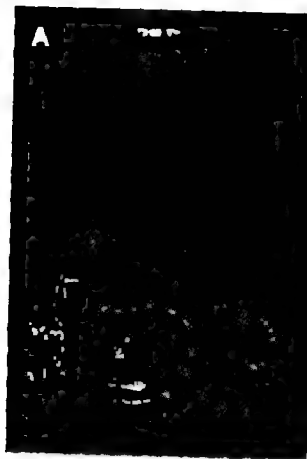


Fig 5 Micrograph under UV light of two otosclerotic stapedes from patients 39 years (A) and 33 years old

(B) ($\times 22$). The foci are quiescent and only a few vascular spaces are lined with newly formed bone

cavités vasculaires, tandis que dans les foyers moins actifs ou quiescents il existe des os néoformés le long des lamelles de quelques ostéons. On ne trouve pas trace de néodépôt osseux dans les étières normaux.

ZUSAMMENFASSUNG

Die Verteilung und die Menge des Knochengewebes das in dem otosklerotischen Herd in 6 Tagen gemacht wurde wurde untersucht in 16 otosklerotischen und 3 nicht-otosklerotischen Steigbügeln, mittels Tetracyclin Markierung. Die relative Menge von fluoreszierenden neu-hergestellten Knochen ist in einer prozentualen Ratio $NA/(NA + VA)$ von neuen Knochen (NA) im Gegensatz zu den vorherigen Knochen (VA) ermittelt worden. In den meisten aktiven Herden wurde eine Schicht von neu-hergestellten Knochen fest um all die in dem Hohlraum befindlichen Blutgefäßen vorgefunden. In den weniger aktiven und ruhenden Herden wurde der neu-hergestellte Knochen nur um den Rand von einigen Osteonen vorgefunden. In den normalen Steigbügeln wurden keine Spuren von neuhergestellten Knochen entdeckt.

REFERENCES

- Alberti, P. W. M. & Tarkenton, J. V. 1963 Stapede otosclerosis: recent histochemical and histological observations. *Laryngoscope* 73 1114.
- Clarke, J. A. 1963 A microscopic study of tetracycline localization in otosclerotic stapes. *J. Laryng.* 79 300.
- Griff, S. 1970. Sulla classificazione istopatologica dell'otosclerosi stapediale: contributo istoradiografico e con marcatura tetraciclica. *Atti della 45^a RI.*
- Frost, H. M. 1961 Human osteoblastic activity II. Measurement of the biological half-life of bone with aid of tetracyclines. *Henry Ford Hosp. Med. Bull.* 9 57.
- Frost, H. M. 1962. Observations on the fundamental nature of otosclerosis. In *Otosclerosis* (ed. H. F. Schubert), pp. 43-6. Churchill, London.
- Frost, H. M., Villanueva, A. R. & Roth, H. 1964. Measurement of bone formation in a 47 year old man by means of tetracycline. *Henry Ford Hosp. Med. Bull.* 8 239.
- García Ibañez, J. L. 1966 Dati preliminari sulla deposizione della tetraciclina nella staffa otosclerotica. *Boll. Soc. It. Biol. Sper.* 43 1660.

- Lee, W. R. 1965 A quantitative microscopic study of bone formation in a normal child and in two children suffering from osteogenesis imperfecta. In *Calcified tissue* (ed. by L. J. Richelle and M. El Dallelmagne), pp. 451-463. Collection des Colloques de l'Université de Liège.
- Milch, R. A., Rall, D. P. & Tobie, J. H. 1958 Fluorescence of tetracycline antibiotics in bone. *J Bone Jt Surg* 40/A 897.
- Roberto, M., Garcia Ibañez, J. & Iurato, S. 1971 Microradiographic studies on the otosclerotic stapes. *Acta Otolaryng* (Stockh.) 72 36.
- 1972. Microhardness testing as a means of analysing the mineralization of the otosclerotic stapes. *Acta Otolaryng* (Stockh.) 73 79.
- Roberto, M. & Morra, F. 1972. Analisi quantitativa dell'accrescimento e del rinnovamento strutturale degli ossicini dell'udito del cane. *Boll Soc It Biol Sper* (in press).
- Röckert, H., Engström, H., Hallén, O., Herberts, O., Lidsén, G., Nordlund, B. & Shea, J. J. 1965 Otosclerosis studied with X-ray microscopy and fluorescence microscopy after administration of tetracyclines. *J Laryng* 79 305.
- Sissons, H. A. & Lee, W. R. 1964 Tetracycline studies of bone turnover. In *Bone and tooth* (ed. by H. J. J. Blackwood), pp. 65-69. Pergamon Press, Oxford.
- van der Stuyt Veer, J., Szencsik, D. & van der Heul, R. O. 1964 Tetracycline labelling of bone in hyperparathyroidism. In *Bone and tooth* (ed. by H. J. J. Blackwood), pp. 85-91. Pergamon Press, Oxford.

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DISCUSSION

Dr Zechner: We hope to show where tetracycline is thought to go. We think that the depolymerized acid mucopolysaccharides are the substances which bind the tetracycline. Those mucopolysaccharides are fairly dense in active otosclerotic foci. These facts were showed by means of coloured slides of post-mortem specimen. They agreed exactly with Dr Iurato's findings.

Dr Engström: The activity of otosclerosis may vary in different areas. One single section through the centre of the stapes may give incomplete information of the stapes as a whole. It would therefore be better to section the footplate "horizontally" and I do believe it is possible to make many sections with the aid of a diamond or special steel knife. I must also congratulate Dr Zechner on his beautiful material, presented in the discussion. It was, however, evident that it was not from the stapes alone.

Dr Iurato (Reply) to Mr Engström: It is really a pity that the diamond saw has a thickness of 300 μ preventing a serial sectioning of the footplate. At maximum, only a few sections can be obtained from a whole footplate.

CLINICAL SIGNIFICANCE AND DIAGNOSTIC VALUE OF AUDITORY BONE THRESHOLD

Some Critical Considerations

O Sala

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Abstract The author surveys the main conditions which lead to a variation of the BC auditory threshold, although lesions of the sensorineural inner ear epithelium may not be shown. The author's criticism of the value commonly assigned to BC threshold impairments is also supported by the results yielded by a scheme of medical therapy in some cases of sensorineural deafness, even lasting since more than 20 years before the treatment. It ensues that a BC threshold elevation should not be considered, as a rule, the expression of an irreversible damage of the inner ear sensory cells. The argument, which holds practical implications mainly from a diagnostic and surgical standpoint, is still the object of experimental and clinical research.

The precise determination of auditory bone conduction (BC) threshold is commonly thought to represent an essential diagnostic clue both in clinical Audiology and Ear Surgery. Several reports on this subject have been published which treat in depth either the methods employed in determining the BC threshold in both normal and pathological conditions, or the difficulties arising from BC masking in asymmetrical hypacusis.

As a rule, BC threshold is assumed as being the sole and true expression of the anatomical and functional features of the inner ear structures and its elevation would represent the very symptom of an unavoidable and irreversible impairment of the inner ear. A critical review of the most recent literature and also some favourable results yielded by a scheme of medical therapy we designed and used in cases of chronic sensorineural deafness

led us to question the semiotic and clinical value generally ascribed to the BC threshold.

As a matter of fact, an elevation of BC auditory threshold may have *extra-labyrinthine origins*. Let us remember for instance: (a) the false labyrinthisation due to the simultaneous blockade of both the oval and round window (Bocca et al., 1968) the inner ear temporary overloading which may be observed either in otosclerotic patients (Carhart's notch) or immediately following a stapedectomy if non-biological prostheses were used (e.g. gelfoam, as we showed by automatic audiometry: Sala & Babighian, 1971) or after a tympanoplasty. Moreover both the AC and BC threshold elevation may be observed when the VIII nerve is impaired (neuromas, tumors of the cerebello-pontine angle) or when cochlear nuclei are somewhat damaged (Fisch 1970).

The BC threshold elevation of *labyrinthine origin* is related to a degenerative involvement of the inner and outer hair-cells. This involvement may be either primitive, or may ensue from some alteration taking place in the supporting structures of the inner ear or in the labyrinthine fluids (Borghesan 1955 Barker 1970). Furthermore, there exist other pathological conditions (some of them already known) which may lead to a BC threshold elevation, although damage to the organ of Corti may not be demonstrated. And namely: (i) vascular stria atrophy (mainly in the apical

turn of the cochlea) (ii) cystic degeneration of the spiral ligament (either at the middle or the apical turn) (iii) primary neural degeneration of the spiral ganglion cells (iv) basilar membrane stiffness, which is followed by a conductive cochlear impairment. This basilar membrane stiffness accounts for some audiological pictures of presbycusis (Schuknecht, 1970; Kerr 1969) and for hypoacusis caused by an endolymphatic hydrops in the initial phases of Menière's disease (Tonndorf 1968).

Each of the four above-mentioned conditions will produce a symmetrical elevation of both the BC and AC threshold, whether or not a damage of the sensorineural epithelium is demonstrable. From an *experimental standpoint* let us remember some recent data. Preliminary studies on the temporary auditory fatigue (TTS), performed by Babighian et al. (1972) with electrophysiological techniques, showed that following intense and prolonged sound stimuli, a peculiar involvement of the superior centers may occur which is added to the peripheral effects of fatigue.

That the higher levels of the auditory system may electrically behave in a different way than the receptor with regard to the same sound stimulus, was similarly shown by Suga & Schlegel (1972), and by Marsh et al. (1961) and Humphrey & Buchwald (1972) mainly in research on habituation. In other words, there is some experimental evidence that an auditory phenomenon which displays a peripheral origin at the audiometric testing may imply central components.

Moreover marked discrepancies were demonstrated between sensorineural hypoacusis and its histological picture. For instance, in cases of permanent auditory fatigue (PTS) electron microscopy studies showed the cell damage at the organ of Corti to be quite disproportionate when contrasted with the audiometric picture (Ward, 1972).

Other elements also support our critical attitude toward the whole problem of BC threshold evaluation.

Spontaneous variations of the BC threshold may be observed in the first phases of Menière's disease (generally accompanying a vertigo attack) in sudden deafness of a viral origin, in sensorineural hypoacusis produced by congenital syphilis (Hahn et al., 1962) in some ototoxic hypoacusis, caused either by salicylate (Myers & Bernstein, 1965), or by quinine (Falbe Hansen, 1941) and in the initial phases of noise-induced hearing loss (Rüedi, 1954). Those threshold variations are likely to be related to biochemical disorders of the ear rather than to sensory cell degeneration.

BC threshold may also vary following *medical or surgical treatment*. In patients treated by *medical therapy* an improvement of BC threshold has been observed in the following cases: in the initial Menière's disease (Beretta & Livan, 1956; De Vincentiis et al. 1964; Giaccal et al., 1968; Klockhoff & Lindblom 1968) in the "idiopathic" sudden deafness (Bosatra & De Stefani, 1963) in sensorineural hearing losses caused by a post-stapedectomy labyrinthitis (Lausse et al., 1971), in some hypoacusis provoked by ototoxic drugs, congenital syphilis, thyroid diseases, collagen diseases (Cody 1971).

Finally we would like to mention some favourable results, yielded by the scheme of medical therapy we devised, which has been applied, throughout 1971 in 70 patients suffering from sensorineural hearing loss of various origin (chronic Menière's disease, ototoxicity haemolytic disease skull injury noise-induced hearing loss, etc.).

As a matter of fact, this scheme hinges on vasodilator drugs, mannitol, heparine, and ACTH anti-bradikinin, anti-serotonin and tissue-oxygenizing drugs are used too. The therapy lasts about 2-3 weeks. It will be pursued at home, throughout 2-3 months, at a reduced dosage. Research upon the action mechanisms of these drugs on the inner ear has been done in our clinic and is still being performed. Some data, which show a pharmacological action on the sensory and/or supporting structures, and

quence a variation of both the AC and BC threshold deserve to be shortly reported.

Automatic audiometry performed immediately before and after mannitol i.v. perfusion showed, in some cases of "chronic" sensorineural deafness, both an improvement of the threshold and a narrowing of the "C" tracing amplitude. In some instances, this phenomenon was strictly dependent on the duration of the perfusion. As a rule these cases showed, at the end of therapy an improvement of both the tonal and vocal threshold, and higher SISI scores than before the treatment (Babighian 1972). Similar DL variations, clearly dependent on mannitol administration, were demonstrated by means of an electrical device which allows to record the stapedial reflex "modulation" amplitude (Babighian & Colletti, 1972). The reflex is elicited by an intense sound and the superimposition of intensity modulated acoustic stimuli. By this technique an objective evaluation of DL changes may be obtained.

The patients we dealt with suffered from a "chronic" sensorineural hearing loss. In addition to amelioration in cases of nuclear deafness and of unknown origin dramatic improvements were observed in some cases of Menière's disease, originating more than 2 years earlier. These cases displayed either a flat or a descending audiometric curve, already stationary the suprathreshold tests being clearly positive.

Details of both the treatment and the results have been given in previous papers (Sala 1971, Sala & Babighian, 1971, 1972). nevertheless, we would like to point out that these un hoped-for results led us to review critically the problems related both to BC auditory threshold measurements and significance, and to its topo-diagnostic and clinical value.

Improvements of BC threshold have been reported following surgery in cases of Menière's disease (Arslan, 1969; Cody 1971; House, 1962), and in decompressive interventions on the cerebellopontine angle (Baker & Christoferson, 1950).

In view of our data, one must therefore be wary about considering the BC threshold as the expression of an irreversible histofunctional impairment of the sensorineural cells. As a matter of fact, the BC threshold elevation may derive from several different inner-ear extralabyrinthine, or perhaps, even nuclear factors, which sometimes may be improved by appropriate medical treatment. The latter also has implications from the pre- and post-surgery standpoint, when we are dealing with patients who need "functional" intervention in the middle ear.

RESUME

L'auteur dans une synthèse critique, prend en considération les conditions qui amènent à une modification du seuil auditif en CO sans que des lésions de l'épithélium neurosensoriel cochléaire puissent être démontrées. Objet de la critique de l'auteur est l'opinion, commune à la plupart des otologistes, que le seuil en CO soit l'expression fidèle des conditions anatomofonctionnelles des éléments neurosensoriels de l'oreille interne, et que son aggravation soit l'expression d'un dommage cochléaire irréversible. Des résultats favorables, obtenus par un schéma personnel de thérapie médicale chez des sujets souffrant par une surdité neurosensorielle « chronique », semblent confirmer cette attitude critique.

Largement, qui présente des implications d'ordre pratique (surtout au point de vue diagnostique et chirurgical), est recouru, à présent, objet de recherches expérimentales et cliniques.

ZUSAMMENFASSUNG

In einer kritischen Synthese berichtet Verf. über die Faktoren, die eine Verleiderung der CO-Hörweite hervorrufen, ohne dass es möglich ist, irgendwelche Schädigungen am neurosensoriellen Scherenschäppel nachzuweisen. Das Objekt der kritischen Betrachtung seitens des Verf. ist die allgemein bei Otologen verbreitete Meinung, dass die CO-Schwelle das exakte Ausdruck der anatomischen und funktionellen Zustände der neurosensoriellen Komponenten des Innenohrs darstellt und dass Verschlechterungen als Anzeichen einer irreversiblen Schädigung angesehen sind. Einige günstige Resultate, die mit einem persönlich ausgearbeiteten medikamentösen Behandlungsschema bei Patienten mit „chronischer“ neurosensorieller Schwerhörigkeit erzielt werden konnten, scheinen diese kritische Einstellung zu rechtfertigen. Dem Thema, das auch praktische Probleme umfasst (besonders in diagnostischer und chirurgischer Hinsicht), werden augenblicklich noch experimentelle und klinische Forschungen gewidmet.

REFERENCES

- Arslan, M. 1969 La méthode de l'induction osmotique dans le traitement de la maladie de Ménière. *Rev Laryng (Bord.)* 90 685
- Babighian, G., Moosheghian, G. & Rupert, A. 1972. Central Manifestations of Auditory Fatigue. An Electrophysiological Study. *Science* (in press).
- Babighian, G. 1972. Sulle moderne tecniche di impedenzometria acustica in otologia. *Valderra* 68 1 — Unpublished data.
- Babighian, G. & Colletti, V. 1972. Un adattatore di impedenza per la registrazione del reperto impedenzometrico. *Minerva Otorinolaring* (in press).
- Baker, G. S. & Christoferson, L. A. 1950. Meningiomas of the cerebellopontine angle: report of a case. *Proc Mayo Clin* 25 549
- Bertini, L. & Livari, M. 1956. *La malattia di Ménière*. Ed. Decca, Milano.
- Bocca, E., Antonelli, A. R., Calero, C., Pugnetaro, O. & Testini, G. P. 1968. La Timpanosclerosi. 56 *Cong Naz Soc Ital ORL (Relaz. Uff.)* Venezia.
- Borghese, E. 1955. *Fisopatologia del canale cocleare. Il Raddio Gruppo Centro-Meridionale Palermo*.
- Bowser, A. & De Stefani, G. B. 1963 The idiopathic sudden deafness, a clinical study. *Acta Otolaryng (Stockh.)*, Suppl. 169
- Bowser, S. K. 1970. The possible importance of the labyrinthine fluids in the pathogenesis of sensorineural deafness. In *Sensorineural hearing loss* (ed. G. E. W. Wolstenholme & J. Knight). Churchill, London.
- Casini, J. Bel, J. Michaux, P. Camu, Y. & Tapon, J. 1971 Chirurgie de l'otospongiose, surdité brusquée après stapedectomie. *Minerva Otorinolaring* 21 11 107
- Cody, T. 1971 Rehabilitation for sensorineural hearing loss. In *Clinical otology An international symposium* (ed. M. Paparella, A. Hohenmann & S. Huff), p. 1 211 C. V. Mosby Co., St. Louis.
- Colletti, V. Unpublished data.
- De Vincentis, J. Bozzi, L. & Pizzichetta, V. 1964 Sulla terapia medica di alcune gravi ipocacosi. *Valderra* 41 65
- Falbe-Hansen, J. 1941 Clinical and experimental histological studies on effects of salicylate and quinine in the ear. *Acta Otolaryng (Stockh.)*, Suppl. 44 1
- Fisch, L. 1970. The selective and differential vulnerability of the auditory system. In *Sensorineural hearing loss* (ed. G. E. W. Wolstenholme & J. Knight). Churchill, London.
- Guacchi, F., Ricci, T. & Franchi, G. C. 1968. Primi risultati di terapia della sordità improvvisa con un farmaco ad azione antiserotoninica. *Boll Ital Otorinolaring* 86 31
- Hahn, R. D., Rodin, P. & Hawkins, H. L. 1962. Treatment of neural deafness with prednisone. *J Chronic Dis* 15 395
- House, W. F. 1962. Subarachnoid shunt for drainage of endolymphatic hydrops: a preliminary report. *Laryngoscope* 72 713
- Humphrey, G. L. & Buchwald, J. S. 1972. Response decrements in the cochlear nucleus of decerebrate cats, during repeated acoustic stimulation. *Science* 175 1488
- Karr, A. G. 1969 The pathology the audiogram and the prognosis in perceptive deafness. *J Laryng* 5 435
- Klockhoff, I. & Lindblom, U. 1968 Glycerol test and Diuretics in Ménière's disease. In *Ménière's disease* (ed. J. L. Pulec), p. 541 Saunders, Philadelphia.
- Marsh, J. T., McCarthy III A., Sheatz, G. & Galambos, R. 1961 Amplitude changes in evoked auditory potentials during habituation and conditioning. *Electroenceph Clin Neurophysiol* 13 224
- Miyers, E. H. & Bernstein, J. M. 1965 Salicylate ototoxicity: a clinical and experimental study. *Arch Otolaryng (Chic.)* 82 483
- Ruedi, L. 1954 Different types and degrees of acoustic trauma by experimental exposure of the human and animal ear to pure tone and noise. *Ann Otol* 63 702
- Schuknecht, H. F. 1970 Functional manifestation of lesions of the sensorineural structures. In *Foundation of modern otology theory* (ed. J. C. Tobias). Academic Press, New York.
- Schuknecht, H. F. 1971 Sensorineural hearing loss—pathologic types and manifestations. In *Clinical otology An international symposium* (ed. M. Paparella, A. Hohenmann & S. Huff), p. 177 C. V. Mosby Co., St. Louis.
- Sala, O. 1971 (Nov.). La terapia medica della sordità. Il Convegno su *La terapia termale in campo otorinolaringologico* Montecatini Terme. In press.
- Sala, O. & Babighian, G. 1973 Automatic versus standard audiometry. *Audiology* 12 21
- 1971 Prospettive di una terapia medica in alcune ipocacosi neurosensoriali. *Minerva Otorinolaring* (in press).
- 1972. Les surdités neurosensorielles chroniques sont-elles irréversibles? *Rev Laryng* 93 513
- Soga, N. & Schlegel, H. 1972. Neural attenuation of responses to emitted sounds in echolocating bats. *Science* 177 82.
- Tomdorf, J. 1968. Pathophysiology of the hearing loss in Ménière's disease. In *Ménière's disease* (ed. J. L. Pulec), p. 375 Saunders, Philadelphia.
- Ward, W. D. 1972. Communication at the *Workshop on ITS* Chicago, Sept. 1 23.

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INFLUENCE OF EXPERIMENTAL HYPOTHYREOSIS ON THE INNER EAR IN RATS

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Abstract Experimental hypothyreosis was induced in four generations of rats. The temporal bones of the fourth generation were prepared for histological studies and the changes in the inner ear are now presented.

This paper is the continuation of the research work undertaken by Gušić and his co-workers in Yugoslavia in the regions where endemic goitres prevail. Clinical studies were made of the changes in the cochleovestibular apparatus of people with endemic goitres. In one of his works Gušić said that "all signs indicating multiple lesions in the region of the pituitary nervous system could be established as the cause of cochleovestibular disorders and not changes in the area of the temporal bone". Such a result of the clinical investigations is not entirely in agreement with the histological findings in temporal bones described in cretins and even in cretinoid dogs by many authors (Alexander 1908, Nager 1917, Manasse, 1909).

This difference prompted us to study the changes in labyrinths of hypothyretic rats. The course of the experiment was prompted by the opinion that changes in the central nervous system and in the process of ossification occur very early during the period of intra-uterine development of a child whose mother has an endemic goitre. Therefore the hypothyreosis was experimentally induced during the intra-uterine development and continued postnatally until the animals were sacrificed.

The experiment was started with 10 healthy female rats of 3 months, weighing 210-250 g. These animals were fed with the standard food to which 0.2 g of Propylthiouracil (PTU, Propocyl Rhenania, Kali-Chemie, Hannover) was added per 100 g. Accordingly the individual daily dosis was about 25 mg. After such treatment over a period of 3 weeks we permitted fertilization. The male rats lived under the same conditions but were fed with standard food only. The fertilized female rats were treated throughout the period of gestation with PTU and this treatment was continued after they had had their litter. The male offspring were removed and the females treated with PTU until they became sexually mature. Fertilization was again permitted by normally fed male rats. After fertilization this first generation was again treated throughout the gestation period with PTU and this was continued even after they had had their young. The experiment was repeated until the fourth generation when we obtained as offspring animals which at the age of 3 months weighed about 40-60 g, had a scanty hairy coat and sluggish movements. This generation was sacrificed and intravital fixation was undertaken. The head was removed and prepared for histological examination.

The second group of animals served for control purposes. These were animals which after birth were immediately put on the previously described treatment with Propyl-



Fig. 1



Fig. 2

Fig. 1 A typical structure of the labyrinthine bony capsule

Fig. 2 Cartilaginous islands in the bony capsule of the labyrinth.

themselves together with their mothers. These animals were also sacrificed when they completed 3 months of life.

The third group of animals also serving as control group consisted of animals fed normally without having been submitted to treatment.

The histological analysis of the labyrinth in

animals of the first group showed that the sensory cells in the labyrinth and the ganglionic cells were normal in their appearance and number. The bone of the labyrinthine capsule did not show the typical lamellar structure; the cellular elements were multiplied as also were the blood vessels (Fig. 1). Cartilaginous tissue remained here and there, especially in



Fig 3

Fig 3 Discontinuity of the epithelium in the region of the prominentia spiralis with the migration of the cells into the ductus cochlearis.

the endosteal layer (Fig. 2) We should like to mention changes in the region of the prominentia spiralis which we were able to establish in each animal of the first group. A discontinuity of the epithelium with migration of the cells into the ductus cochlearis was involved here (Fig. 3)

For the time being the interpretation of this finding is purely speculative. The finding of cartilaginous islands in the bony capsule of the labyrinth has sometimes been seen in rats without any treatment or disease.

In animals of the second and the third group the previously mentioned changes could not be established. The findings were entirely normal.

RÉSUMÉ

Les os temporeux de la quatrième génération de rats hypothyroïdiques ont été examinés histologiquement. Les altérations dans l'oreille interne sont décrites.

ZUSAMMENFASSUNG

In Ratten wurde eine experimentelle Hypothyreose durch vier Generationen erhalten. Die Schläfenbeine der vierten Generation wurden für histologische

Untersuchung vorbereitet und die Veränderungen im Innenohr werden präsentiert.

REFERENCES

- Alexander G 1908 Des Gehörorgan der Kröten. *Arch Ohrenheilk* 78 34
- Griff, B. 1957 Über die Kochleovestibulären Störungen bei endemischer Struma. *Pract Otorhinolaryng* (Basel) 19 531
- 1963 Die Rolle des Otologen in der Bekämpfung der endemischen Struma. *Monat Ohrenheilk* 1 225
- 1964 Über die Kochleovestibulären Veränderungen beim endemischen Kropf. 10 Symposium der Deutschen Gesellschaft für Endokrinologie über Schilddrüsenhormone und Körperperipherie Regulation der Schilddrüsenfunktion. Seite 207 209
- Griff, B. Femenč, B. & Konč-Carnežni, V. 1963 Mittelohrschleimhautveränderungen bei experimenteller Dysfunktion der Schilddrüse. *Acta Otol* (Stockh.) 69 281
- Manasse, P. 1909 Über kongenitale Taubstummheit und Struma. *Z. Ohrenheilk. Krankheiten Larynx* 58 105
- Nager F. R. 1917 Zur Anatomie der endemischen Taubstummheit (mit einem Neurofibrin- und Schneckenpendel). *Z. Ohrenheilk. Krankheiten Larynx* 73 349
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FREEZE DRIED BONE AS A MEATAL IMPLANT

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Abstract. As part of a programme to improve the operative results in surgery for congenital meatal atresia, we have studied the fate of freeze-dried homograft bone in dogs when implanted as an isotopic graft. Histological studies were made of the graft reaction at regular intervals up to 2 years. Osteogenic activity was monitored by tetracycline fluoroscopy. Regional lymph nodes were examined for evidence of graft rejection. There was no homograft reaction and the graft acted as a scaffold for new bone formation.

Most otologists will agree that the commonest complication of operations for congenital atresia of the ear is that of stenosis of the newly fashioned meatus. Indeed, this was found in approximately 23% of 181 operations performed in the Radcliffe Infirmary Oxford, since 1958. In an attempt to overcome this problem it was decided to examine the possible value of a two-stage procedure in which the first step would consist of burying a bony homograft meatus in situ and closing the skin completely over it.

The present paper is an account of the findings obtained in dogs subjected to the first stage.

MATERIAL AND METHOD

The graft consisted of bony meatus alone with no soft tissue attached. Grafts were taken under clean but not sterile conditions from cadaver heads which had been refrigerated at 2°C for 48 hours following sacrifice.

Sterilisation was achieved by immersion in 2 ethylene oxide for 12 hours. Ethylene

oxide is a highly volatile fluid which boils at 10.7°C and sterilisation was carried out in a fume cabinet at 4°C. Aerobic and anaerobic cultures from the grafts and containers showed no growth on nutrient broth or Robertson's meat medium, and there were no wound infections in the experimental animals. Following sterilisation the grafts were placed in Pyrex glass containers and snap frozen to -79°C in a mixture of dry ice and ethyl alcohol prior to freeze drying in a conventional freeze drying apparatus. Drying was completed when the vacuum pump reached 0.01 mmHg indicating that sublimation of water vapour from the graft had ceased (Fig. 1).

The grafts were then stored under vacuum and reconstituted with normal saline before implantation.

The hosts were male dogs aged 6 months to 2 years and litter mates were not used as donors and hosts. After excision of the pinna the outer portion of the host meatus was enlarged to accept the graft, which was wedged into position after suitable trimming with a cutting burr.

The graft reaction was studied at regular intervals up to 2 years. The meatuses and surrounding bone were decalcified in 5% nitric acid, paraffin embedded and sectioned in the sagittal plane. Tetracycline-labelled specimens were embedded in plastic, and sections viewed under ultraviolet light. Lymph nodes from both sides of the neck were examined for evidence of an immune response.

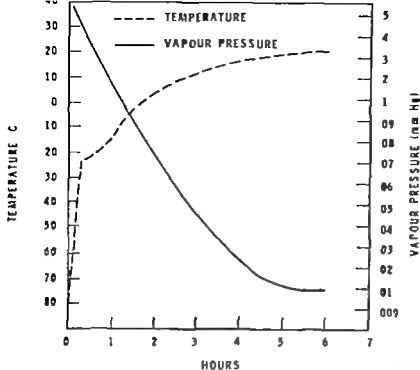


Fig 1 Graph of average dry time for grafts of between 3-4 g dried weight. Vapour pressure was measured with a Fin vacuum gauge.

RESULTS

During the first month there was considerable activity at the surfaces of the graft and it was separated by a layer of granulation tissue apart from small pressure points where the graft had been wedged into position (Fig 2). The host surface showed evidence of osteogenic activity with new woven or non-lamellar bone growth into the granulation tissue (Fig 3).

This process was further advanced at 2 months and active osteoblasts lined the host surface. The graft showed early attack by multinuclear osteoblasts with numerous Howship's lacunae, but this did not appear to persist. At 2 months the graft surface showed little activity. There was no evidence of new bone formation at this stage. Lymph nodes examined at 2 months showed reactive hyperplasia only.

The 3-month and 16-month sections showed new bone formation on the surface of the graft, both in the intramedullary and free ends.

Fig 2 Low-power view to show free portion of graft in relation to host bone. The graft was covered by a polythene obturator was used.



Fig 3 Surface of host bone showing marked osteogenic activity with new woven bone formation occurring in the adjacent granulation tissue.

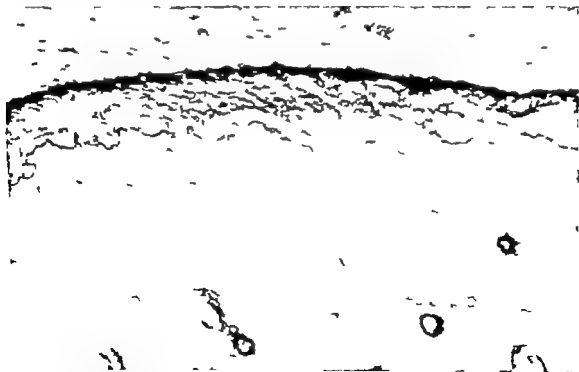


Fig 4 Three-month graft consisting mainly of dead bone but new layer of living host bone lines the graft surface.

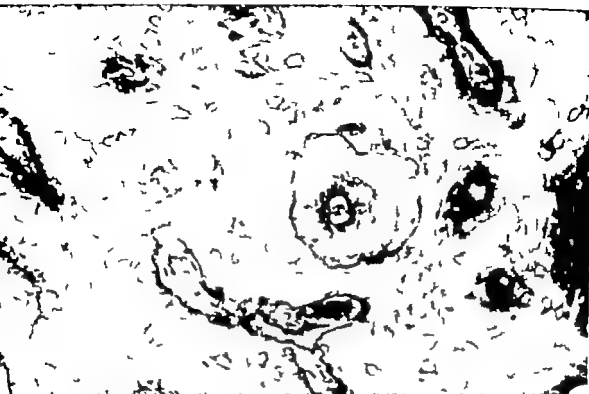


Fig. 5 Body of 6-month graft showing an island of new bone formation around a vascular channel.



Fig. 6 Tetracycline-labelled new bone seen on the graft surface of a section taken at 3 months.

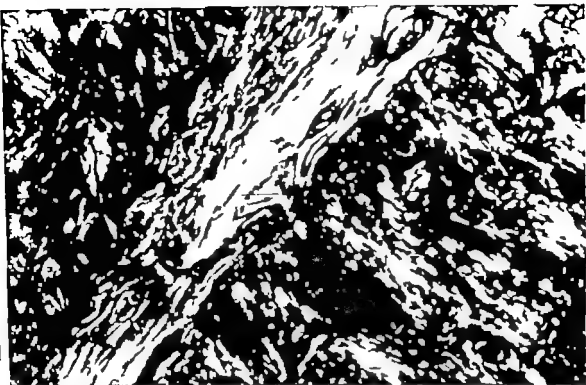


Fig 7 Polarised light view of graft-host fusion at 1 year showing interweaving of bone collagen.

portions (Fig. 4). Small islands of new bone were seen in the body of the graft, surrounding the vascular channels with cement lines radiating from them (Fig. 5). The graft host junction was recognised by the firm apposition of living and dead bone. Tetracycline-labelled new bone was seen on the surface of the graft when sections were viewed under UV light (Fig. 6).

The 1 year and 2 year sections showed a continuous process of graft fusion with host bone. Polarised light examination revealed intermingling of graft and host bone collagen (Fig. 7). The graft underwent a process of creeping substitution, that is, gradual replacement of dead by living bone. At 2 years it remained as a firm bony meatus with new bone lamellae surrounding residual islands of dead bone (Fig. 8).

CONCLUSION

As a result of these experiments in dogs it was felt justifiable to use a similar method in hu-

man patients. Although the patients were children, an adult size meatus was implanted this was done in the same way as the dog experiments. The first patient had the first stage

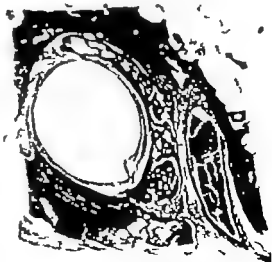


Fig 8 Two-year graft showing a firm bony meatus of good thickness. Scattered islands of dead bone were still present in the body of the graft.

done in May 1970. Her second stage was performed a year later at which the obturator was removed, the meatus Thiersch grafted, and local flaps fashioned the line the meatal entrance. The meatus remains stable and of good size. We believe that homograft meatuses can be used to advantage in certain patients with congenital atresia, and it is planned to report at a later stage on our patients who have received homograft meatuses.

RÉSUMÉ

Dans le cadre d'une étude expérimentale dont le but était d'améliorer les résultats du traitement chirurgical des atresies congénitales du conduit auditif externe, nous avons examiné le sort d'homogreffes osseuses autotopiques hypophysectées chez le chien. La réaction des greffes fut suivie par des contrôles histologiques effectués à intervalles réguliers pendant une durée de 2 ans. L'activité ostéogénique fut mesurée par fluoroscopie à la tétracycline. Les glandes lymphatiques régionales furent examinées afin d'y mettre en évidence des signes de rejet. Il n'y a pas eu de réaction aux homogreffes, les greffons servant de support à l'ostéogénèse.

ZUSAMMENFASSUNG

Als Teil eines Programmes zur Verbesserung der chirurgischen Ergebnisse in der Behandlung kongenitaler Atresien des äusseren Gehörorgans, wurde das Los gefroren-getrockneter autotopischen homogenen Knochentransplantate in Hunden studiert. Histologische Untersuchungen der Transplantate wurden in regelmässigen Abständen während 2 Jahren ausgeführt. Die osteogenetische Aktivität wurde mittels Tetracyclin-Fluoroskopie ausgemessen. Regionale Lymphknoten wurden für Zeichen von Ausstossungen der Transplantate untersucht. Es war keine Reaktion zu den homogenen Transplantaten vorhanden, und

die Transplantate dienten als Rahmen für die Knocheneubildung.

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DISCUSSION

W. F. House: (1) Why use polyethylene dioxide and freeze drying instead of 70% alcohol which has been found satisfactory in homograft ossicles? (2) Do the grafts line well with skin or does stenosis occur from granulation in the lumen of the grafted external canal?

I. Friedmann: Aseptic new bone formation is an interesting experiment is of great importance. Is the newly formed bone originating in the bone or is external auditory meatus? What has happened to the squamous epithelium of the meatus? Could the new bone formation lead to the formation of exostosis or the complete osseous obliteration of the canal?

J. L. W. Wright (Reply) to Mr House and W. Friedmann: Work by Burwell and others has shown that freeze-dried bone will foster new bone formation whereas boiled, methylated and deproteinized bone does not. That is, the former seems to encourage osteoblast formation by release of bone inductors from the organic substrate.

Woodruff has also stated that freeze drying is the most effective way to impair the antigenicity of large bone grafts. Our human grafts are considerably larger than incus bones and we thought therefore that this was an important factor.

The grafts were implanted with a small polyethylene obturator which stimulated an early well differentiated fibrous bed, thus reducing the number of well differentiated connective tissue cells available to stick the graft surface. In dogs we noticed that middle ear epithelium grew out to line the meatus, which showed no tendency to exostosis formation during the period of study. In human patients we have implanted the meatuses as part of a two-stage procedure. At the second stage, 9 months later the meatus was found to be lined with a thin layer of fibrous tissue which was suitable for a Thiersch graft. The middle ear mucosa had closed off the middle ear space, forcing the medial layer of a future tympanic membrane.

SYMMETRICAL HEARING LOSS IN BRAIN STEM LESIONS

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Abstract. An examination of bilateral deafness resulting from proven lesions of the brain stem has shown that the pure tone audiograms at the two ears are invariably identical. This finding has interesting theoretical implications concerning the tonotopic organisation of the auditory pathways in the brain stem which will be discussed.

Certain facts about the clinical effects of lesions of the central pathways of the cochleas are well known. Thus, for example removal of one cerebral hemisphere in animals has only a trifling effect upon hearing as judged by the pure tone audiogram and indeed we ourselves have been unable to detect any significant difference in the hearing level at the two ears of patients subjected to hemispherectomy.

The explanation of this seems to be that above the level of the cochlear nuclei there is a substantial division of the cochlear pathways as shown in Fig. 1 so that the surviving hemisphere remains supplied with impulses from both cochleas.

Now this argument may well be applied to a unilateral lesion at any point above the decussation and it follows therefore that a unilateral retro-cochlear hearing loss can only result from a lesion of the cochlear nuclei on one side or the nerve fibres peripheral to them but not from more central unilateral lesions.

By the same token a brain stem lesion above the level of the cochlear nuclei can only give rise to deafness if it involves the cochlear pathways bilaterally and in this event there should be deafness at both ears. We know

however from the work of Rose et al. (1960, 1963) and others that there is present in the brain stem a tonotopic organisation of the auditory fibres at all levels.

Adopting the same argument as before this leads to interesting speculation concerning the character of the hearing loss at the two ears since it may be presumed that central bilateral deafness can only occur when fibres subserving identical frequency bands on both sides of the brain stem are involved. In this event bilateral deafness will result which must of necessity be symmetrical in respect of its frequency distribution at the two ears.

This is illustrated schematically in Fig. 2 in which are shown the two central cochlear pathways left and right within the brain stem each with its tonotopic arrangement of fibres. The enclosed areas on each side represent lesions resulting for example from plaques due to multiple sclerosis. Although as will be seen the involvement on one side is considerable, deafness can only result from those frequency regions which are common to both sides indicated by the solid shading.

With these considerations in mind we have reviewed some 500 patients with brain stem lesions who have been examined in the Neuro-otological Clinic at Queen Square, London. As was to be expected deafness was not a common finding nevertheless in those patients in whom deafness was present and temporally related to the brain stem disorder bilateral symmetrical hearing loss was found

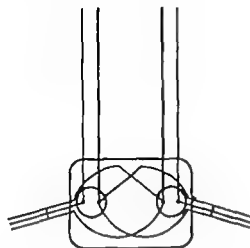


Fig. 1 Schematic diagram of cochlear nerves and central pathways.

to be the rule. This is exemplified in Fig. 3 in which are shown the audiograms from 9 patients all with confirmed brain stem lesions including 3 cases of intramedullary glioma, 3 cases of cerebellar glioma involving the brain stem 2 cases of degenerative disease involving cerebellum and brain stem and 1 of diffuse vascular disease. The findings from autopsy operation or other investigations are summarised below.

B, a woman of 25 was under the care of Dr Gautier-Smith, and had had progressive deafness, ataxia of gait and clumsiness of arms for 15 years, together with intermittent regurgitation of fluids. Three paternal aunts and paternal grandmother were deaf since childhood. Posterior fossa decompression 11 years ago had demonstrated no abnormality but was followed by improvement.

Neurological examination revealed poor palatal movement and depressed gag reflex. Tongue movements were slow and clumsy. She had marked cerebellar ataxia, worse in left arm and leg with left hyperreflexia. Myelography showed a dilated fourth ventricle. A.E.G. showed cerebellar atrophy.

Neuro-otological abnormalities included a bilateral symmetrical perceptive deafness for all frequencies within the range 200–8 000 Hz;

very severe for the higher tones. Loudness recruitment was complete as assessed by loudness discomfort levels. She had good toleration of amplified speech. Tone decay was present bilaterally at 1 000 2 000 and 4 000 Hz. First-degree spontaneous nystagmus of rebound type was present to left and right and upwards. Following movements and optokinetic responses were deranged, caloric responses exaggerated. There was positional nystagmus of central type.

Diagnosis: Familial degenerative process involving cerebellum and its vestibular and ocular connections including the cochlear pathways within the brain stem.

E. P., a woman of 50, was under the care of Dr Goody and complained of difficulty in focusing her eyes for 5 months with diplopia on lateral and downward gaze. She was unable to look up.

Neurological examination revealed limited ocular elevation and convergence, impairment of tone, power and co-ordination of left arm. A.E.G. showed a mass in the region of the quadrigeminal plate producing obstruction of the upper third of the aqueduct.

Neuro-otological abnormalities included slight bilateral symmetrical deafness in the range 200–8 000 Hz. Loudness recruitment was complete as assessed by loudness discomfort levels. She had unsteadiness of gait. Convergence was absent and she was unable to look upwards and had first-degree vestibular nystagmus to right. Optokinetic responses showed directional preponderance to right and were present upwards but absent downwards. Caloric responses were exaggerated with directional preponderance to right.

Progress. Torkildsen's operation was performed with temporary relief of symptoms and she had a course of deep X-ray therapy. A year later she was admitted with recurrence of symptoms and ventriculography showed more severe hydrocephalus. She deteriorated and died. Autopsy was refused.

Diagnosis. Glioma in region of posterior

end of third ventricle involving the right vestibular and both cochlear pathways in the mid-brain.

L. H., a woman of 45 was under the care of Mr McKissock, and had suffered for 4 years with severe frontal headaches and attacks of giddiness, nausea and vomiting. For 16 months she had difficulty in hearing and bilateral tinnitus unrelated to headache or vertigo.

Neurological examination revealed bilateral papilloedema with defective vision, worse in right than left eye. X rays of skull showed a crescent of linear calcification above and behind pineal region. Ventriculography demonstrated a tumour 3 cm diameter producing a filling defect of the third ventricle. Appearances suggested a globular tumour overlying the quadrigeminal plate with calcification of its posterior wall.

Neuro-otological abnormalities included a mild bilateral symmetrical perceptive deafness in the frequency range 200-4 000 Hz. She had some ataxia of gait deviating to right. Caloric responses showed slight directional preponderance to right.

Progress. Partial excision of an inoperable tumour was undertaken but patient died within 24 hours. Autopsy showed tumour arising from quadrigemina. Pathological report: Oligodendroglioma.

Diagnosis: Quadrigeminal plate glioma.

G W., a man of 67 under the care of Professor Gilliatt. Eight years ago he had severe headache followed by unconsciousness after which he had vertigo and vomiting and mental confusion since when he has remained unsteady with mild deafness. Two years ago he became depressed. An A.E.G. 8 months ago showed communicating hydrocephalus but since this he developed urgency and incontinence of micturition and impaired memory.

Neurological examination revealed mild dementia, monotonous voice and expressionless face and mild ataxia of limbs. B.P. 220/115

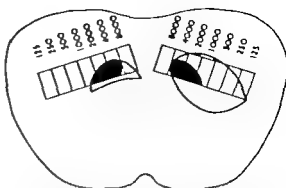


Fig. 2. Schematic diagram of left and right central cochlear pathways showing tonotopic arrangement of fibres. Shaded areas represent deafness resulting from asymmetric bilateral lesions.

Electrocardiography showed left ventricular hypertrophy C.S.F. 100 mg Protein/ml. A.E.G. showed hydrocephalus with air over surface of hemispheres suggesting subarachnoid block.

Neuro-otological abnormalities included a progressively severe bilateral symmetrical perceptive deafness in the frequency range 200-8 000 Hz. Loudness recruitment complete as assessed by loudness discomfort levels. He had ataxia of stance and gait, falling or veering to right. Optokinetic nystagmus was deranged in character. Electro-nystagmographic records showed first-degree vestibular nystagmus to left and right abolished by eye closure and darkness.

Progress. A right V A. Holter Rickham shunt operation was performed with resulting improvement in symptoms.

Diagnosis: Subarachnoid haemorrhage with resulting communicating hydrocephalus in a hypertensive patient following vascular accident involving the right vestibular and the cochlear neurones bilaterally at a high level in the brain stem.

J M., a man of 59 was under the care of Dr Gooddy and complained of headache and feeling drunk for the past 10 days. He was unsteady on his feet and giddy on rising from a sitting posture.

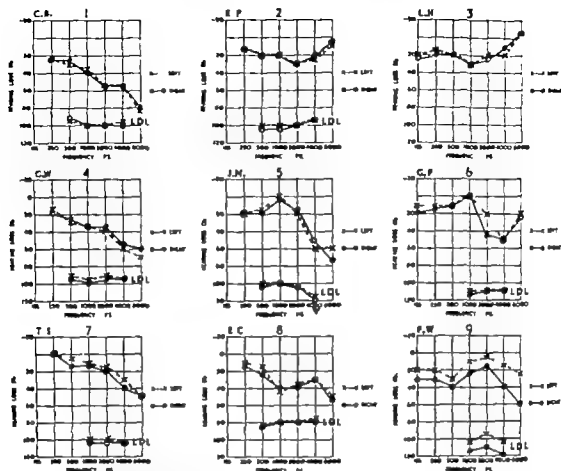


Fig. 3. Audiograms of 9 patients with confirmed brain lesions: Cerebellar and Brain Stem Atrophy 1 and 8. Intramedullary Glioma. 2, 3 and 9. Diffuse

Vascular Disease: 4 Cerebellar Glioma involving Brain Stem 5, 6 and 7.

Neurological examination revealed that he had severe nausea and vomiting on sitting up from a lying position.

Neuro-otological abnormalities included a symmetrical bilateral perceptive deafness in the frequency range 4 000–8 000 Hz. Loudness recruitment was present for the frequencies 500–2 000 Hz as assessed by loudness discomfort levels. He was very unsteady on standing, falling to right and was unable to walk. He had first-degree vestibular nystagmus to the left upward gaze was impaired. There was positional nystagmus of central type in the left lateral position. Caloric responses were exaggerated showing directional preponderance to left combined with right canal paresis.

Progress. Air encephalography revealed

herniated cerebellar tonsils. At subsequent operation a cystic tumour was excised from right cerebellar hemisphere. The dura was under greatly increased tension. There was immediate post-operative improvement in the clinical condition but patient died 2 months later from *Haemophilus Meningitis*.

Pathology Haemangioblastoma.

Diagnosis. Right cystic cerebellar glioma with secondary brain stem involvement implicating cerebello-vestibular cerebello-ocular and right vestibular pathways together with the intramedullary cochlear neurones bilaterally.

G.F. a man of 59 was under the care of Mr McKissock and had 1 month's history

of occipital headache, difficulty with speech, giddiness and loss of power in right arm.

Neurological examination revealed cerebellar dysarthria with ataxia of right arm and leg. C.S.F. Protein 500 mg/100 ml.

Neuro-otological abnormalities included bilateral perceptive deafness substantially limited to the frequency range 2 000–8 000 Hz. Loudness recruitment present as assessed by loudness discomfort levels. He stood and walked very unsteadily falling to right. Slight first-degree vestibular nystagmus to left was present. He had positional nystagmus of central type and derangement of optokinetic nystagmus which was worse to right than left. The left caloric responses were reduced.

Progress. A right cystic cerebellar tumour was excised which extended into the fourth ventricle. Good recovery.

Pathology. Haemangioblastoma.

Diagnosis. Right cerebellar glioma involving cerebello-vestibular vestibular and cochlear pathways bilaterally within the brain stem.

T.S., a man of 58, was under the care of Dr Ross Russell, and had throbbing morning headaches for 14 months in the right temporo-occipital region, aggravated by coughing, laughing or straining. He also had progressive weakness and unsteadiness of legs, deviating to right on walking. He had noticed right-sided deafness in past 6 months.

Neurological examination revealed spastic quadriparesis, worse in legs, with bilateral cerebellar signs worse right than left. Ventriculography showed dilatation of lateral and third ventricles with displacement to left of fourth ventricle.

Neuro-otological abnormalities included a severely severe bilateral perceptive deafness in the range 200–8 000 Hz. Loudness recruitment was complete as assessed by loudness discomfort levels. He was more easily pushed to right than left on standing and deviated to right on walking with eyes closed. Upward gaze and convergence were limited

and optokinetic responses sluggish. Electro-nystagmographic records showed much saccadic movement in darkness. Caloric responses showed directional preponderance to left.

Progress. Vascular tumour lying posterolaterally in right cerebellar hemisphere and fed by posterior inferior cerebellar artery was removed with good recovery including improved eye movements.

Pathology. Haemangioblastoma.

Diagnosis. Right cerebellar glioma displacing brain stem and involving cerebello-ocular right vestibular and intramedullary cochlear pathways bilaterally.

E.C., a woman of 67 was under the care of Dr Gooddy and had had four brief episodes of dizziness 9 months ago. For the past 4 months she had progressive unsteadiness of gait with clumsiness of hands.

Neurological examination revealed trunkal ataxia with cerebellar inco-ordination of all four limbs particularly legs and very brisk deep tendon reflexes. The right plantar response was extensor.

Neuro-otological abnormalities included bilateral symmetrical perceptive deafness in the frequency range 200–8 000 Hz, more severe for the higher tones. Loudness recruitment was complete as assessed by loudness discomfort levels. She had ataxia of gait and directional preponderance to left of caloric responses.

Progress. Balance deteriorated steadily. She developed positional nystagmus of central type and died 2 years later. Autopsy showed generalised cerebellar atrophy with a pons diminished in size. Microscopically cells of pontine nuclei were diminished in numbers with fibrous gliosis, axons thin with faintly staining myelin sheaths.

Diagnosis. Olivo-ponto-cerebellar atrophy involving cerebello-vestibular and central cochlear pathways bilaterally.

F.W., a woman of 48, was under the care of Mr McKissock, and had had intermittent

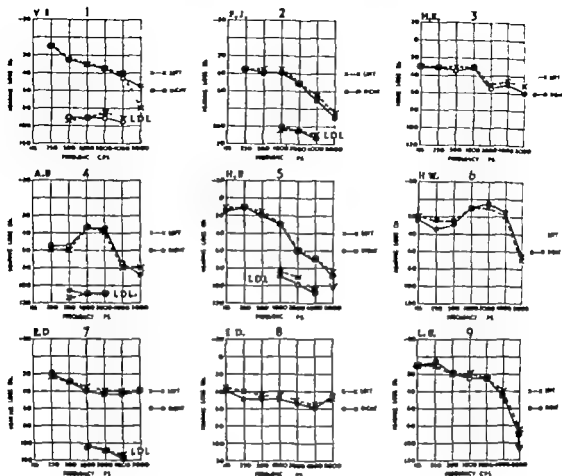


Fig. 4 Audiotograms of 9 patients with brain stem lesions diagnosed on clinical findings: Multiple Sclerosis, 1; Vascular Disease, 2, 3, 4, 5, 7 and 9; basilar medullary Metastases, 6; Pellagra, 8.

rocks. 1 Vascular Disease 2, 3, 4, 5, 7 and 9, basilar medullary Metastases; 6. Pellagra. 8.

discharge from right ear for years. Two months ago she had developed diplopia, headaches, numbness of left side of face with loss of taste ataxia and stiffness of limbs, particularly on right side.

Neurological examination revealed dysarthria, diplopia in all directions of gaze, with left trigeminal loss both motor and sensory. She had right lower motor neurone facial weakness and weakness of left side of palate and all four limbs with pyramidal signs and inco-ordination with marked weakness and impaired superficial sensation on right side. Ventriculography demonstrated dilated lateral and third ventricles with posterior displacement of fourth ventricle suggesting a pontine tumour. A limited A.E.G. demonstrated pontine enlargement. Vertebral arteriography

confirmed a space-occupying lesion in the pons more on left side with upward tentorial herniation.

Neuro-otological abnormalities included a slight bilateral symmetrical perceptive deafness. This was complicated on the right side by a superimposed conductive deafness due to an active right chronic suppurative otitis media. Loudness recruitment was complete bilaterally as assessed by loudness discomfort levels, the loudness discomfort level in the right being elevated only by the amount of the conductive deafness. There was an abductor paresis of the left vocal cord. Spontaneous nystagmus was present to left and right and upwards with positional nystagmus of central type and absent optokinetic responses to right and downwards. Caloric responses were

grossly exaggerated with directional preponderance to left.

Diagnosis. Intramedullary glioma involving right vestibular neurones and left nucleus ambiguus, the cerebello-vestibular and central cochlear pathways being involved bilaterally within the brain stem.

With the exception of Case F W in which the difference in hearing levels at the two ears could be attributed to a conductive defect the bone conduction audiograms confirmed the hearing loss in the remainder to be sensorineural in origin. The symmetry of the hearing loss at the two ears is self-evident. In addition it will be seen that the Loudness Discomfort Levels are within the normal range and therefore indicate the presence of complete recruitment. This is a finding of some significance and we shall return to it later.

In Fig. 4 are shown the audiograms of a selection of patients with brain stem lesions unconfirmed by operation. Summaries of the clinical findings in each are as follows.

V E. a woman of 60 was under the care of Professor Gilliat, and had had progressive difficulty in walking for 15 years, urgency of micturition, stress incontinence, blurred vision, diplopia and, more recently parasthesiae involving fingers of right hand and feet.

Neurological examination revealed bilateral intention tremor and exaggeration of reflexes in upper limbs, weakness of lower limbs with ankle clonus and hyper-reflexia. Vibration sense was absent in right leg.

Neuro-otological abnormalities included severe ataxia of stance and gait, spontaneous nystagmus to left and right of rebound type, inhibited in darkness and abolished by eye closure. Following movements were broken up and optokinetic responses sluggish. Caloric responses showed directional preponderance to left combined with right canal paresis.

Diagnosis. Multiple Sclerosis with cerebellar and pyramidal involvement and lesions of

right vestibular and both cochlear pathways within the brain stem.

F J., a man aged 66 was under the care of Professor Gilliat, and, 3 months ago had sudden loss of consciousness with paralysis of legs, vertigo and vomiting and had been temporarily deaf. The legs gradually recovered. He had throbbing, occipital headache 20 years ago.

Neurological examination of fundi revealed arteriolar narrowing. He had a right temporal hemianopia and constriction of left temporal field. There was bilateral heel/shin ataxia.

Neuro-otological abnormalities included bilateral symmetrical perceptive deafness, more severe for the higher tones, associated with loudness recruitment as assessed by the loudness discomfort levels. He had ataxia of gait, reduction of left caloric responses and inconsistent deviations in both directions of assessment of the visual vertical and horizontal.

Diagnosis: Vascular insufficiency with cerebellar and posterior cerebral lesions and recent vascular accident involving the central cochlear pathways and left vestibular neurones within the brain stem.

M. K. was a woman of 69 under the care of Dr Critchley. She had deteriorated mentally and physically over the past year and suffered from falling attacks without vertigo or loss of consciousness, for 6 months.

Neurological examination revealed mild dysphasia and slurring dysarthria. All tendon reflexes were exaggerated and plantar responses extensor.

Neuro-otological abnormalities included bilateral perceptive deafness and ataxia of stance and gait. There was defective upward gaze and left vestibular nystagmus to left and right. Optokinetic responses were defective in the vertical plane. Caloric responses showed slight reduction of the left reactions.

Diagnosis. Diffuse cerebro-vascular disease affecting cortex, anterior mid brain pathways

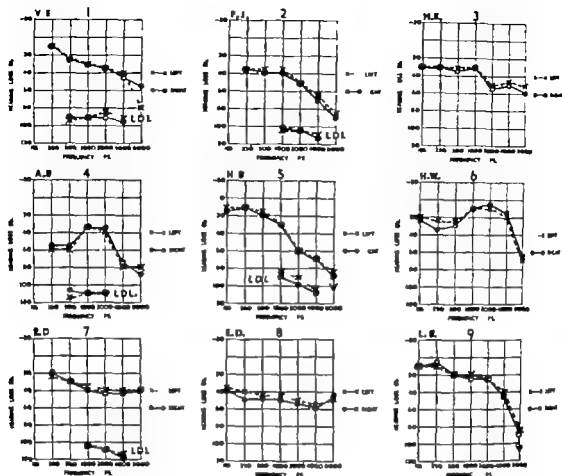


Fig. 4 Audiograms of 9 patients with brain stem lesions diagnosed on clinical findings: Multiple Sclerosis: 1 Vascular Disease: 2, 3, 4, 5, 7 and 9. Medullary Metastases: 6. Pellagra: 8.

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discharge from right ear for years. Two months ago she had developed diplopia, head aches, numbness of left side of face with loss of taste, ataxia and stiffness of limbs, particularly on right side.

Neurological examination revealed dysarthria, diplopia in all directions of gaze, with left trigeminal loss both motor and sensory. She had right lower motor neurone facial weakness and weakness of left side of palate and all four limbs with pyramidal signs and inco-ordination with marked weakness and impaired superficial sensation on right side. Ventriculography demonstrated dilated lateral and third ventricles with posterior displacement of fourth ventricle suggesting a pontine tumour. A limited A.E.G. demonstrated pontine enlargement. Vertebral arteriography

confirmed a space-occupying lesion in the pons more on left side with upward tentorial herniation.

Neuro-otological abnormalities included a slight bilateral symmetrical perceptive deafness. This was complicated on the right side by a superimposed conductive deafness due to an active right chronic suppurative otitis media. Loudness recruitment was complete bilaterally as assessed by loudness discomfort levels, the loudness discomfort level in the right being elevated only by the amount of the conductive deafness. There was an abductor paresis of the left vocal cord. Spontaneous nystagmus was present to left and right and upwards with positional nystagmus of central type and absent optokinetic responses to right and downwards. Caloric responses were

weeks ago this had been associated with vertigo, vomiting and weakness and cramp in the legs. She had a feeling of being tilted to the left and had diplopia, slurred speech, headache and weakness and clumsiness of left hand. All symptoms were improving.

Neurological examination revealed impaired sensation to pain and temperature over right cheek, right side of trunk and right lower limb together with weakness and incoordination of left leg. B.P. 220/75. There was evidence of coronary infarction.

Neuro-otological abnormalities included a bilateral symmetrical perceptive deafness with loudness recruitment present as assessed by loudness discomfort levels. She was ataxic on standing and walking. The caloric responses were a little exaggerated with relative reduction of the left reactions.

Diagnosis. Hypertension with recent brain stem vascular accident.

E.D., a woman of 43 was under the care of Professor Dent, and gave an 8 year history of episodes of lassitude, depression, deafness and weight loss in which her skin had become dry dark and thick. She had diarrhoea and the difficulty in hearing was worse at times of attacks. Marked improvement in symptoms on each occasion had resulted from Nicotinic Acid therapy.

Neurological examination revealed dark pigmentation of skin with hyperkeratosis of ankles and dorsum of hands. She was mentally confused and disorientated with spastic quadriceps and extensor plantar responses, ankle and patellar clonus.

Neuro-otological abnormalities included bilateral symmetrical perceptive deafness. The caloric responses were exaggerated bilaterally with normal pattern. There was patchy pigmentation of palate.

Diagnosis. Pellagra involving intramedullary cerebello-vestibular and cochlear pathways bilaterally.

L.B., a woman of 64 was under the care of Dr Williams, and had had difficulty in walking during the past year. Her hearing had been deteriorating some 4-5 years.

Neurological examination revealed multiple neurofibromata of skin with exaggerated tendon reflexes and extensor plantar responses. She had complete achlorhydria. B.P. 240/120. E.E.G. showed episodic activity considered to be a result of brain stem ischaemia.

Neuro-otological abnormalities included bilateral symmetrical perceptive deafness in the range 500-8 000 Hz, severe at 8 000 Hz. She had ataxia of stance and was unable to walk with the eyes closed. There was positional nystagmus of central type and exaggeration of caloric responses with normal pattern.

Diagnosis. Diffuse organic defect involving the cerebello-vestibular pathways and cochlear neurones bilaterally within the brain stem likely to be due to vascular insufficiency associated with her hypertension.

Once again a feature of all the above cases is the symmetry of the hearing loss at the two ears and in addition, according to the loudness discomfort levels, loudness recruitment seems to be the rule.

DISCUSSION

It can be argued that the hypothesis we have presented to account for this symmetry is unnecessary since our findings might equally well be explained in terms of symmetry of the lesion of the kind commonly encountered in congenital hearing loss. Deafness due to kernicterus is a case in point.

The audiograms of six typical cases are shown in Fig. 5. In these the lesion is known to involve the cochlear nuclei themselves (Bertrand, 1946; Crabtree & Gerrard, 1950; Dublin, 1951; Gerrard, 1952) and on this account in accordance with our hypothesis it must be presumed that the symmetry of the hearing loss reflects a similar symmetry of the lesion. While it cannot be denied that this explanation needs to be taken into account

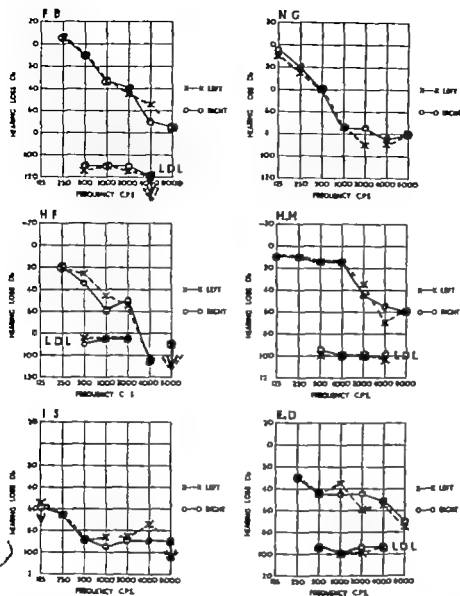


Fig. 5. Audiograms of 6 patients with deafness due to Kernicterus.

considering the cases in our series the probability of such a wide variety of brain stem lesions giving rise to symmetrical bilateral degeneration of the cochlear nuclei would seem to be a remote one.

A second possibility that needs to be considered is that the deafness resulted from either a symmetrical bilateral nerve fibre or cochlear lesion. The former can certainly be ruled out since it will be recalled that loudness recruitment was a constant finding in all of our cases. As to the latter in all the cases reported here it will be recalled that the deaf-

ness was shown to be a related symptom of the disorder. The occurrence of a coincidental cochlear lesion therefore would imply a statistical improbability. Nevertheless the consistent finding in our cases of loudness recruitment, a phenomenon which has long been taken to be characteristic of and restricted to cochlear lesions, remains to be explained. More recent studies including those of Libin (1969) and Carhart (1967) incline to the view that loudness recruitment is a feature of brain stem disorders. In this respect the occurrence of recruitment in all of the cases of kern-

icterus reported here in which the test was carried out is of some significance. Blakeley (1959), Flottorp et al. (1957) and Matkin (1965) have all reported similar findings. In addition there is now a substantial body of histopathological evidence based upon the examination of temporal bones (Wolf 1956, Crabtree & Gerrard, 1950; Gerrard, 1952, Goodhill, 1956) that end organ involvement is conspicuously absent in kernicterus. In this respect it is of some significance that in all the cases we have examined the caloric responses have been normal. This would suggest that the lesion is at a level where the cochlear and vestibular fibres have separated. It must be inferred therefore that the presence of recruitment in these cases results from derangements of the cochlear nuclei.

This occurrence of loudness recruitment in brain stem lesions has obvious and important theoretical implications outside the scope of this discussion. Suffice it to say here that it would seem to imply either that the mechanism of recruitment in brain stem lesions differs from that commonly attributed to end organ lesions or else some revision of the latter is called for.

Our studies are continuing and we hope in due course to be able to support our hypothesis with histopathological evidence. In this event the recognition of symmetrical hearing loss exhibiting loudness recruitment in patients with known or suspected brain stem lesions could well be a localising sign of some significance.

The cases referred to in this paper included the following Hospital Numbers (N.H.Q.S.): A65504 A9455 A36450, A43008, 53465 52163 A61507 A10241 A40289 A26277 A16195 A10901 A47037 A9327 A25473 78050, A22855 A22243 A40395 81906, A51643 A52715 A24393

ACKNOWLEDGMENTS

Thanks are due to the Consultants of the National Hospital, Queen Square and elsewhere who have kindly referred their cases for examination.

RÉSUMÉ

L'examen de patients souffrant d'ictères confirmés du tronc cérébral, avec hyponcousse bilatérale a montré que l'afectus auditif tonale est pratiquement toujours identique pour les deux oreilles. Cette constatation a des implications théoriques intéressantes en ce qui concerne l'organisation somatotopique des voies auditives dans le tronc cérébral. Elles seront le sujet d'une discussion.

ZUSAMMENFASSUNG

Untersuchungen von Fällen von bilateralem Gehörsmangel durch erwiesene Mittelhirnherden verursacht, haben gezeigt, dass die Reinton-Audiogrammen der zwei Ohren stets identisch sind. Dieser Befund hat interessante theoretische Konsequenzen über die tonotopische Organisation der Gehörverbindungen im Mittelhirn: diese werden weiter besprochen.

REFERENCES

- Bertrand, L. 1946. Lésions du système nerveux central dans deux cas d'ictère du nouveau-né. *Rev. Hém. (Par.)* 1: 339.
- Blakeley R. W. 1959. Erythroblastosis and hearing loss responses of siblicolts to tests of cochlear function. *J. Speech Hearing Res.* 2: 5.
- Carhart R. 1967. Audiological tests: Questions and speculations. In *Deafness in childhood* (ed. F. McConnell & P. H. Ward), p. 229. Vanderbilt University Press.
- Crabtree N. & Gerrard, J. 1950. Perceptive deafness associated with severe neonatal jaundice: Report of 16 cases. *J. Laryng.* 64: 442.
- Dubin, W. 1931. Neurological lesions of erythroblastosis fetalis in relation to nuclear deafness. *Amer. J. Clin. Path.* 21: 935.
- Flottorp G. D., Morley E. & Skatvedt M. 1957. The localization of hearing impairment in siblicolts. *Acta Otolaryng. (Stockh.)* 48: 404.
- Gerrard, J. 1952. Nuclear jaundice and deafness. *J. Laryng.* 66: 39.
- Goodhill, V. 1956. Clinical pathological aspects of kernicteric nuclear deafness. *J. Speech Hearing Dis.* 21: 407.
- 1967. Auditory pathway lesions resulting from Rh incompatibility. In *Deafness in childhood* (ed. F. McConnell & P. H. Ward), p. 215. Vanderbilt University Press.
- Liden G. 1969. The scope and applications of current audiometric tests. *J. Laryng.* 83: 507.
- Matkin, N. D. 1965. *Audiological patterns characterizing hearing impairments due to Rh incompatibility*. Ph.D. dissertation, Northeastern University.
- Morales-Garcia, C. & Hood, J. D. 1972. Tone decay in neuro-otological diagnosis. *Arch. Otolaryng. (Chic.)* 96: 231.
- Morales-Garcia, C. & Poole, J. P. 1971. Masked

speech audiometry in cortical deafness. *Acta Otolaryng* (Stockh.) 74: 307

- Rose, J. E., Galambos, R. & Hughes, J. R. 1960. Organisation of frequency sensitive neurons in the cochlear nuclear complex of the cat. In *Neural mechanisms of the auditory and vestibular systems* (ed. G. L. Rasmussen & W. F. Windle), chap. 9. C. C. Thomas, Springfield, Ill.
- Rose, J. E., Greenwood, E. P., Goldberg, J. M. & Hind, J. E. 1963. Some discharge characteristics of single neurons in the inferior colliculus of the cat. I. Tonotopical organization, relative of spike counts to tone intensity and firing patterns of single elements. *J. Neurophysiol.* 26: 294
- Wolf, D. 1956. (quoted by Goodhill, 1967).

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DISCUSSION

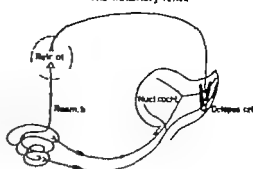
P. Pieloux. J'ai été très intéressé, car j'ai observé moi-même plusieurs malades atteints de lésions du tronc cérébral, du point de vue auditif j'ai constaté l'atteinte symétrique. Cependant je voudrais me permettre quelques réserves l'étiologie paraît jouer un rôle dans cet aspect symptomatique. Par exemple si ce dernier se retrouve de façon presque constante dans les tumeurs du IV^e ventricule il paraît exceptionnel dans les gliomes du tronc cérébral. Cette constatation amène à penser que l'un des éléments jouant un rôle dans l'atteinte auditive symétrique pourrait être la situation anatomique de la lésion mélanche au niveau des noyaux cochléaires ou de la connexion.

R. Hinckeliff. I would congratulate Miss Dix on tackling a problem which in its more general aspect has puzzled audiologists for many years. Le the frequent symmetry of tonal threshold audiograms. This applies not only to the types of lesions which she has studied but also to lesions of the internal and middle ear. Of course there is also the possibility of double coincidental lesions of which, I am sure, she is aware.

G. Liden. I want to compliment Miss Dix for her very interesting paper. Poole and Hood stated recently that an increased level of loudness discomfort (LDL) was a good indicator of retrocochlear deafness. The cases of Miss Dix seemed to have a normal LDL. Were any supplementary audiological tests done to exclude the possibility of simultaneous lesion in the cochlea?

C. R. Pfaltz. Lesions of the cochlear nuclei are causing integrational disturbances of the process of hearing. I should like to ask Miss Dix if she has carried out any integrational hearing tests such as Bocca's or Feldmann's test to find further evidence for the pres-

The inhibitory reflex



ence of a central, i.e. integrational, deafness in the group of patients.

J. M. Aron. Very shortly I hope to report some observations we have made on the VIII nerve response in one case of brain stem degeneration, in kernicterus cases and also in retrocochlear lesions. These observations could help in the discussion of Miss Dix' presentation and support her hypothesis, particularly about the possibility of a central origin for loudness recruitment in some instances. In the case of brain stem degeneration we recorded absolutely normal responses showing no recruiting phenomenon. In the kernicterus cases as well as in most of the retrocochlear disorders we always observe very abnormal, distorted patterns which indicate that the lesion must be also partly peripheral. Also in the kernicterus cases the recordings are quite similar symmetric, on both sides.

J. G. Hall. As to kernicterus, you will see in my thesis from 1964 that there is a great loss of nerve cells in the cochlear nuclei following kernicterus. The bilirubin hampers the oxygen carrying ability of the haemoglobin so the cells disintegrate, either because of the toxic effect of the bilirubin or of asphyxia.

As to the possibility of a central recruitment, a paper was presented on this topic by Liden, Engström and myself in Gothenburg, this year. Due to new neuroanatomical findings, the nervous pathways inducing a "central recruitment" may be described. Dr Kirsten Owen described the octopus cells in the dorsal part of the dorsoventral cochlear nucleus. These cells are so situated that incoming impulses must pass through their area. Their afferents pass to the new olivary bundle where the efferent bundle of Rasmussen originates, carrying inhibitive impulses (Fig. 1). This short reflex consisting of three neurons has a constant tonus, inhibiting unwanted impulses. Now if this tonus should be disturbed, due to a disease or due to a tumour somewhere along its course the inhibition would fall, the impulses pass undisturbed, and you would have a "central recruitment".

M. Dix (Reply) to Mr Pieloux. Dr Dix said that bilateral symmetrical deafness had certainly been encountered in patients with proven neurological abnormalities rostral to the level of the cochlear nuclei as for example in cases E. H. and L. H. with phenos-

involving the quadrigeminal plate and G W with communicating hydrocephalus and electrocystagmographic evidence of a high brain stem lesion. Although there was no clinical evidence that these lesions extended distally to involve the IV ventricle or cochlear nuclei this possibility could not of course be excluded.

Mr Hbacht//: Dr Dix pointed out that examinations of the ears and tests of bone conduction had been entirely normal apart from the single case of F W. This finding taken together with the fact that the complaint of or demonstration of symmetrical deafness had been related to central disorders of such a wide variety would make the likelihood of a coincidental cochlear lesion a statistical improbability. This view was fortified by the demonstration of bilateral symmetrical deafness in keraticus, a condition in which the lesion was known to involve the brain stem and in which normal cochleas had been demonstrated histologically.

Mr Liden: Dr Dix pointed out that what had been stated by Hood and Poole (1966) was that with an elevated or absent Loudness Discomfort Level "the lesion has an extremely high probability of the order of 9 cases out of 10 of being in the nerve fibre". No reference was made to the findings in brain stem

lesions. Tone decay tests carried out more recently on a number of patients with brain stem lesions had demonstrated abnormal results in a significant proportion whereas with cochlear lesions the presence of tone decay was exceptional. (Morales-Garcia and Hood, 1972.) As an example, in the case of C.B. in the present study tone decay was present at all frequencies tested, this amounted to 35 decibels at 4 000 Hz.

Mr Pfaltz: Dr Dix said that although no tests of integrational deafness had been carried out on the group of patients in the present study further work on a group of 15 patients with brain stem lesions had shown that their masked speech scores were significantly lower than those of normal subjects but no correlation had yet been established with respect to the level or lateralisation of the lesion (Morales-Garcia and Poole, 1972).

Mr Hall: Dr Dix thanked him for his kind and helpful remarks. The neuro-anatomical mechanisms which he described would purport to be most appropriate and would provide just such an explanation as we had been looking for to account for central recruitment.

A COMPARISON BETWEEN MIDDLE EAR MUSCLE REFLEX THRESHOLDS FOR BONE AND AIR-CONDUCTED PURE TONES

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Abstract. The middle-ear muscle reflex threshold for bone-conducted pure tones (250-6 000 Hz) was compared with the corresponding threshold for air-conducted tones. In normal-hearing subjects a significant difference (5-25 dB) was found. Possible causes for this discrepancy were studied.

It is well known that acoustic stimulation of high intensity produces bilateral changes in the acoustic impedance of the ear. These impedance changes are brought about by reflex contractions of the middle-ear muscles. The middle-ear muscle reflex threshold for air-conducted pure tones is 70-90 dB sensation level (Jepsen 1955). This has been confirmed by numerous investigators.

The corresponding reflex threshold for bone-conducted pure tones has, until now, only been studied in a few patients with Menière's syndrome in which the findings were difficult to evaluate (Djupesland, 1964).

The main purpose of the present investigation was to determine the middle-ear muscle reflex threshold (from now on referred to as "the reflex threshold") for bone-conducted pure tones in normal-hearing subjects and to compare it with the threshold for air-conducted tones.

MATERIAL AND METHOD

Our material consisted of 7 subjects with normal hearing in both ears (3 female, aged 19-26 years, and 4 male, aged 27-46 years) and 7 subjects with normal hearing in one ear the

contralateral being totally deaf (2 female, aged 23-59 years, and 5 male, aged 16-35 years). In all subjects the contralateral ear and the tympanic membrane had a normal appearance.

The reflex contractions of the middle-ear muscles were studied by means of a Madsen Electro-Acoustic Impedance Bridge (Model ZO 70). Using bridge sensitivity setting 3, the reflex threshold was defined as the smallest intensity giving approximately 10% of maximal obtainable impedance change.

The middle-ear pressure was measured as described by Terkildsen & Thomsen (1959) and found to be normal in all cases. In all ears, even in the totally deaf ones, changes in the acoustic impedance of the ear could be elicited by touching the skin around the opening of the ipsilateral and the contralateral auditory meatus with cotton wool (tactile stimulation). These findings indicate normal middle-ear function (Djupesland, 1967, 1969).

The subject was positioned on an examination table in a soundproof and almost anechoic chamber. The head was placed on a cushion so that the mastoid process was in a horizontal position. Through an opening in the cushion, the ear was connected to the impedance bridge (Fig. 1). In the unilaterally deaf subjects, the impedance bridge was always connected to the deaf ear.

The signals, giving air and bone-conducted pure tones, were delivered from a Peter



Fig. 1 Subject placed on the examination table with Mini Shaker (\rightarrow) applied to the right mastoid process and the left ear connected (\Rightarrow) to the impedance bridge.

Audiometer type SPD 5. The duration of each signal was approximately 1.5 sec. The signals were either directly delivered to a earphone (Telephonics TDH 39) or amplified and delivered to a Mini Shaker (Bruel & Kjaer Type 4810) or a loudspeaker (Tandberg BK 165). A block diagram of the equipment is shown in Fig. 2. The audiometer was calibrated according to the ISO 1964 standard. The harmonic distortion and the maximum output of the Mini Shaker were determined. Results are shown in Table I.

The sound-pressure level generated by the loudspeaker was measured in the ear of the subjects with a Brüel & Kjaer Precision Sound Level Meter Type 2203 equipped with an octave filter set, Type 1613.

The Mini Shaker was placed on the mastoid process facing upwards, without contact with the pinna (Fig. 1). The pinna was bent somewhat in a forward direction, using adhesive tape. (The ear canal was not occluded.)

Hearing threshold and reflex measurements

Using the earphone and the Mini Shaker the hearing thresholds for the frequencies 250, 500, 1 000, 2 000, 3 000, 4 000 and 6 000 Hz were measured by the method of limits. In addition the reflex thresholds for air and bone-conducted pure tones were determined for the same frequencies. All measurements were performed at three different times for the normal hearing subjects, and in one session only for the unilaterally deaf persons.

Loudness balance test

Since the middle ear muscle reflex in man is loudness governed (Metz, 1952; Ross 1968; Flottorp et al. 1971 and others), we thought it of interest to perform loudness balance test between air and bone-conducted sounds according to the method of Fowler (1936).

In the normal-hearing subjects the impedance bridge was disconnected, keeping the rubber plug and the two polyethylene tubes in the ear canal. The polyethylene tubes were connected to a TDH 39 earphone, using an adapter.

In the unilaterally deaf subjects, the loudness balance test refers only to one ear. The earphone and the Mini Shaker could not be placed simultaneously on the same side. Therefore, the earphone was replaced by a loudspeaker hanging 35 cm above the entrance of the ear canal. The hearing threshold measurements, both for air and bone-conducted sounds, were repeated in order to relate all loudness levels to threshold values.

Control experiments

To make sure that the observed impedance changes were caused by middle-ear muscle contractions and not by artefacts (for in-

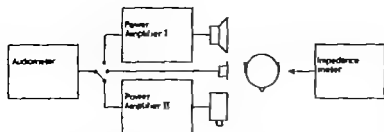


Fig. 2. Block diagram of the equipment showing the 3 different types of sound sources: loudspeaker, earphone and Mini Shaker stimulating one ear. Impedance meter connected in the opposite ear.

stance, interference between the stimulus tone and the probe tone of the Impedance bridge) some supplementary experiments were carried out. Three totally deaf persons with normal tympanic membranes and 3 with tympano-sclerosis on one side and normal hearing on the other side served this purpose. In the deaf subjects, ear canal irrigations with water at temperatures of 44, 30 and 4 degrees Centigrade respectively were not followed by nystagmus, indicating that the labyrinths were out of function. However tactile stimulation elicited reflex contractions of the middle-ear muscles on both sides, indicating normal middle-ear function.

Using air- and bone-conducted pure tones of maximal obtainable intensity (Table I) no impedance changes were observed in any of these subjects. These findings indicate that the acoustically elicited impedance changes, observed in the present investigation were caused by genuine reflex contractions of the middle-ear muscles and not by artefacts.

RESULTS

The reflex thresholds for air- and bone-conducted pure tones in normal-hearing persons are presented in Figs. 3 and 4. In Fig. 3 the hearing as well as the reflex thresholds are presented relative to normal hearing thresholds for air (ISO 1964) and bone-conducted pure tones (DAI 1972).

The values in Fig. 4 are related to the hearing thresholds measured individually just before the determination of the reflex thresholds.

As shown in Figs. 3 and 4 a considerable

difference between reflex thresholds for air- and bone-conducted pure tones was found, especially in the low frequency range. This difference amounted to 5–25 dB.

In the high frequency range, middle-ear muscle reflex could not be elicited in all subjects for bone-conducted stimuli, even when maximum intensity (Table 1) was used (circled symbols 2 000–6 000 Hz).

At high intensity levels of the frequencies 250, 500 and 1 000 Hz, the Mini Shaker in all cases produced a tingling sensation localized in the area below the bone vibrator. A distinct threshold for the tingling sensation was found except for the high frequency range (2 000–6 000 Hz) where no such sensation was reported. The threshold of the tingling sensation coincided with the reflex threshold.

The results of the loudness balance test are plotted in Fig. 4. The rate of loudness increase as a function of sensation level was approximately the same for air- and bone-conducted pure tones.

The results for the unilaterally deaf subjects were practically identical with those reported for the normal-hearing group.

DISCUSSION

The results have shown that in normal-hearing subjects there is a considerable difference in reflex thresholds for air- and bone-conducted pure tones. The middle-ear muscle reflex in man, elicited by air-conducted pure tones, seems to be loudness governed (Mitz 1952, Ewertsen et al 1958, Djupesland & Flottorp, 1970, Flottorp et al., 1971). Therefore, the observed difference indicates that

Table I. Harmonic distortion and maximum level of Mini Shaker

Frequency (Hz)	Level of harmonic components in dB re 1 mV			Max. output in dB re bone conduction threshold (DAI)
	1 H	2 H	3 H	
250	30	28	2	68
500	49	27	1	73
1 000	41	-12	-34	81
2 000	26	-38	-34	75
3 000	17	-40	-35	65
4 000	12	-30	-12	65
6 000	11	-25		70

The reference zero for bone-conduction (DAI) in this Table is based upon an investigation carried out at our institute (Det Audiologiske Institut, DAI) not yet published, however proposed as a preliminary "standard" until such has been established by ISO.

the reflex mechanism for bone-conducted pure tones cannot be loudness governed. This is further supported by the results of the loudness balance experiments, which show approximately the same rate of loudness increase for air- and bone-conducted pure tones.

Among the pronounced differences between the two types of stimuli the following may be mentioned.

(a) The bone vibrator (Mini Shaker) was in direct contact with the skin, thereby adding the possibility of tactile stimulation of the reflex mechanism (Djupesland, 1967) whereas the air-conducted tones do not produce any tactile sensation.

(b) The Mini Shaker stimulates both cochleae with almost the same intensity whereas the air-conducted tones only affect one ear.

In order to investigate whether the difference in reflex eliciting mechanisms might be due to one or both of the above-mentioned modes of stimulation, experiments were conducted, excluding the tactile possibility by means of anaesthesia, and binaural summation by using unilaterally deaf subjects.

(c) The skin between the Mini Shaker and the mastoid process was anaesthetized by injections of 5 ml of Xylocain® 1% in 3 subjects. The reflex threshold was found to be

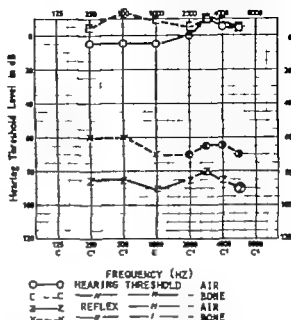


Fig. 3 Hearing and reflex threshold for air and bone-conducted tones in normal-hearing persons.

the same as before the injection. Therefore, the possibility of a tactile-elicited reflex can be excluded. Further support for this exclusion is the fact that in the previously mentioned totally deaf subjects, maximum stimulation with the Mini Shaker caused no impedance change.

(b) Seven subjects with normal hearing in one ear and a deaf contralateral ear showed no significant difference in reflex thresholds from the seven normal-hearing subjects. Thus a possible binaural summation effect can be excluded.

The subjects reported that the tonal sensation changed at and above the tingling sensation threshold. We therefore initiated an investigation of the content of aural harmonics in normal-hearing subjects for bone-conducted pure tones. The method of "best beats" was used (Opheim & Flottorp, 1955).

Our results so far seem to indicate that unusual overload of the inner ear starts at the tingling sensation threshold. Therefore, a possible difference in inner ear distortion

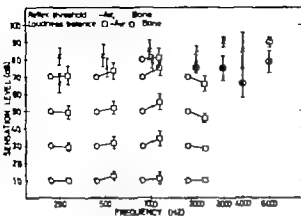


Fig. 4 "Reflex threshold and equal loudness for air and bone-conducted tones in normal-hearing persons.

for air and bone-conducted pure tones may be responsible for the difference in reflex thresholds.

RÉSUMÉ

Le seuil d'excitation du réflexe musculaire de l'oreille moyenne sollicité par des sons purs (250-6000 Hz) conduits par voie osseuse est comparé avec le seuil correspondant pour des sons conduits par l'air. Chez des sujets à audition normale, on a trouvé une différence significative (5-25 dB). Les causes possibles de cette différence ont été recherchées.

ZUSAMMENFASSUNG

Die Mittelohrmuskelreflexschwelle bei knöchengeleiteten, reinen Tönen (250-6000 Hz) wurde verglichen mit der entsprechenden Schwelle bei luftgeleiteten Tönen. Bei Normalhörigen wurde ein signifikanter Unterschied (5-25 dB) festgestellt. Mögliche Ursachen dieses Unterschieds wurden studiert.

REFERENCES

- Djupesland, H. 1964 Mechanical component to deafness in Meniere's disease. *Acta Otolaryng* (Stockh.), Suppl. 188 206.
 — 1967 *Contractions of the tympanic muscles in man*. Thesis. Universitetsforlaget Oslo Norway
 — 1969 Use of impedance indicator in diagnosis of middle ear pathology. *Int Audiol* 8 570.
 Djupesland, G & Flottorp G 1970. Correlation between the Fowler loudness balance test, the Metz recruitment test and the Flottorp-Opheim's aural harmonic test in various types of hearing impairment. *Int Audiol* 9 156.
 Ewertsen, H. W., Filling, S., Terkildsen K. & Thom-

- sen, K. A. 1958 Comparative recruitment testing. *Acta Otolaryng* (Stockh.) Suppl. 140 116.
 Fowler E. P. 1936. A method for the early detection of otosclerosis, a study of sounds well above the threshold. *Arch Otolaryng* (Chic.) 24 731.
 Flottorp, G., Djupesland, G & Wether F 1971. The acoustic stapedius reflex in relation to critical bandwidth. *J Acoust Soc Amer* 49 457.
 I.S.O. standard 1964 *ISO Recommendation R 39* "Standard reference zero for the calibration pure-tone audiometers".
 Jepsen, O 1955 *Studies on the acoustic stapedius reflex in man*. Measurements of the acoustic impedance of the tympanic membrane in normal individuals and in patients with peripheral facial palsy. Thesis. Universitetsforlaget, Aarhus, Denmark.
 Metz, O 1952. Threshold of reflex contraction: muscles of middle ear and recruitment of low tones. *Arch Otolaryng* (Chic.) 55 336.
 Opheim, O & Flottorp G 1955 The aural harmonic in normal and pathological hearing. *Acta Otolaryng* (Stockh.) 45 513.
 Ross S. 1968 On the relation between the acoustic reflex and loudness. *J Acoust Soc Amer* 43, 768.
 Terkildsen, K. & Thomsen, K. A. 1959 The influence of pressure variations on the impedance of the human ear drum. *J Laryng* 73 409.

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DISCUSSION

G Liden. I agree that it is a great advantage to be able to elicit the stapedial reflex at about 30 dB lower level than with a conventional technique. However bone vibrators of commercially available audiometers do not seem to be able to do so. I wonder if Mr Djupesland has excluded the possibility of physical interference between the stimulus and the carrier tones?

R. Hinchcliffe. Mr Djupesland has mentioned most of the possible sources of error but I wonder whether he could comment on the possibility that there is acoustic interference with the operation of the impedance bridge. As he knows, one of the modes of bone transmission is via the air in the external acoustic meatus—and this would also be on the air fitted with the impedance bridge.

G Djupesland (Reply) to Mr Liden: An ordinary bone vibrator cannot be used to measure the middle ear muscle reflex threshold in normal-hearing persons because of limited output and non-linearity at high intensity levels.

To Mr Hinchcliffe. The problem concerning recording artefacts for example interference between the stimulus tone and the probe tone (carrier tone)

of the impedance bridge) was studied using 3 totally deaf persons with normal tympanic membranes and 3 patients with tympanosclerosis on one side and normal hearing on the other side. In the present paper these control experiments are reported (not mentioned

during my presentation). The results indicated that the acoustically elicited impedance changes, observed in the present investigation, were caused by genuine reflex contractions of the middle-ear muscles and not by artefacts.

THE EFFECTS OF ETHACRYNIC ACID UPON THE COCHLEAR ENDOLYMPH AND STRIA VASCULARIS

A Preliminary Report

S. K. Bosher C. Smith and R. L. Warren

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Abstract. The normal endocochlear potential was rapidly replaced by a negative anodic-type potential after ethacrynic acid administration (60 mg/kg) in the rat. Recovery at first proceeded quickly but became very much slower at 30 min. However the endolymphatic chemical composition remained unaltered until 35 min when progressive increase in the sodium and decrease in the potassium concentrations occurred, followed at 1 hour by a gradual return towards normal. Thus the drug probably causes transient inhibition of the stria enzymes and then abnormalities in membrane permeability. The characteristic accumulation of inter-cellular fluid in the stria vascularis was associated with and seemed to arise from these permeability changes. In addition, the findings strongly support the concept that the endocochlear potential has two components, a positive secretion potential and a negative potassium diffusion potential.

The discovery early in its clinical use, that both transient and permanent deafness could be produced by the diuretic agent, ethacrynic acid, has been followed by a number of investigations into the possible mechanism of its action upon the cochlea. Of particular prominence in the initial period were those studies showing the cochlear microphonic responses to be depressed and often abolished by the drug under a wide variety of conditions (Mathog et al. 1970). In addition, electron microscopic examination has considerably increased our knowledge of the effects induced for not only has the occurrence of the degeneration of the basal outer hair cells described first in human material (Matz et al.,

1969) been confirmed, but the presence of morphological abnormalities in the stria vascularis has also been revealed (Quick & Devall, 1970).

This discovery of stria involvement led naturally to the suggestion that alterations in endolymph composition might have an important role in the pathogenesis of the deafness and, latterly direct experimental evidence about such a possibility has been obtained (Cohn et al. 1971 Silverstein & Yates, 1971). The rather diverse results, however are difficult to interpret and nothing is known about possible disorders of the endolymphatic anion content. Furthermore, no information was available concerning the endocochlear potential, a vital factor in determining the ionic gradients, until the recent report by Prazma and his colleagues (1972). These authors were, in fact, able to demonstrate the presence of marked variations in this potential and to correlate these variations with the effects observed upon the other cochlear potentials, but they did not themselves investigate the ionic concentrations of the inner ear fluids in their animals.

The present investigation was therefore undertaken to determine the extent of the changes in the endocochlear potential and the endolymphatic constitution following ethacrynic acid administration in a single species and

to correlate these, as far as possible, with the ultrastructural anomalies produced in the stria vascularis, in order to determine the nature of the drug's deleterious action upon the endolymph system.

MATERIAL AND METHODS

Adult white Wistar rats, body weight 200 g, were used for the experiments and anaesthetized with Urethane (1.66 mg/kg body weight by intra-peritoneal injection) Ethacrynic acid, as sodium ethacrynate (12.5 mg/ml solution), was administered by intravenous injection into the left femoral vein in a dose of 60 mg/kg body weight, the injection time being 25–30 seconds.

The acoustic stimuli for the cochlear microphonic responses, 2 kHz, tone pulses of 100 ms duration, were produced by means of a Brüel & Kjær type 1024 sine random generator and delivered through a Brüel & Kjær type 4216 constant sound pressure source placed 6 inches from the animal's intact left external ear. The stimulus level was adjusted to give a signal magnitude of 90 dB (re 2×10^{-5} N/m²), as recorded by a half-inch condenser microphone placed in the position of the animal's tympanic membrane and measured by a Brüel & Kjær type 2606 measuring amplifier. The cochlear microphonic potentials were recorded using a stainless steel wire electrode placed on the bony margin of the round window after the left bulla had been opened in the post-auricular region. The potentials were amplified by means of a Tektronix 3A9 unit, displayed on a Tektronix 565 oscilloscope and photographed with a Tektronix C27 camera.

Details of the experimental procedures for the measurement of the endocochlear potential and the collection of samples of the inner ear fluids, together with the analytical methods employed will be found in two previous publications (Bosher & Warren, 1968, 1971) to which reference should be made.

Animals selected for ultrastructural exami-

nation were sacrificed by decapitation 2 hours post-injection, after the removal of endolymph from the left cochlea. The temporal bones were removed and the cochleae fixed for 2 hours by either phosphate-buffered (pH 7.2–7.4) 1% osmium tetroxide or 3% glutaraldehyde at 4°C. The glutaraldehyde-fixed material was post-fixed in 1% osmium tetroxide. The specimens were next embedded in Epon and subsequently divided into appropriate segments by means of a jeweller's lathe. Sections of the middle turn were prepared using a Cambridge Huxley ultra-microtome, stained with uranyl acetate and lead citrate, and examined in a Philips EM 300 electron microscope.

RESULTS

The cochlear potentials

The dose of ethacrynic acid administered was selected because it was found to be the smallest one which, in the rat, produced gross reduction of the cochlear microphonic potentials (to 10% in approximately 15 min) and particular attention was directed to the early changes since these were of the greatest interest.

With regard to the endocochlear potential, striking effects were invariably found, as in the examples shown in Fig. 1. After a short latent period of 2–3 min, the magnitude of the potential declined at first slowly but then extremely rapidly reaching a minimum of -42 to -50 mV at about 16 min. Thereafter recovery initially was rapid but its rate soon decreased considerably and minor interanimal variations became apparent. However at 60 min the potential had attained approximately $+30$ mV in all the animals examined and the rate of potential increase became uniformly low for at least the next hour. The general pattern of these changes is in good agreement with the experimental records published by Prazma and his co-workers (1972), although these authors do not comment upon the biphasic recovery process.

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1969) been confirmed, but the presence of morphological abnormalities in the stria vascularis has also been revealed (Quick & Devall, 1970).

This discovery of stria involvement led usually to the suggestion that alterations in endolymph composition might have an important role in the pathogenesis of the deafness and, latterly, direct experimental evidence about such a possibility has been obtained (Cohn et al. 1971; Silverstein & Yekta 1971). The rather diverse results, however, are difficult to interpret and nothing is known about possible disorders of the endolymphatic anion content. Furthermore, no information was available concerning the endocochlear potential, a vital factor in determining the ionic gradients, until the recent report by Prazma and his colleagues (1972). These authors were in fact, able to demonstrate the presence of marked variations in this potential and to correlate these variations with the effects observed upon the other cochlear potentials, but they did not themselves investigate the ionic concentrations of the inner ear fluid in their animals.

The present investigation was therefore undertaken to determine the extent of the changes in the endocochlear potential and the endolymphatic constitution following ethacrynic acid administration in a single species and

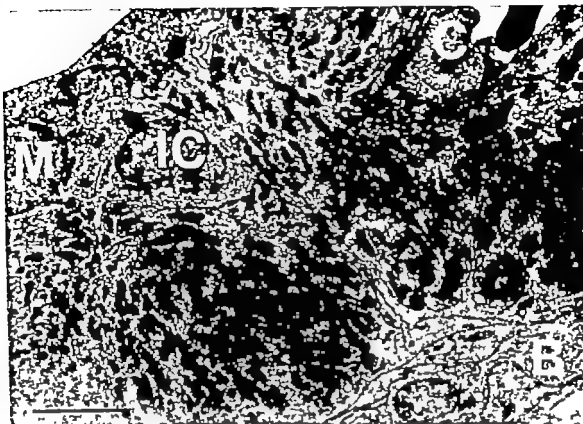


Fig. 3. Stria vascularis 2 hours post-injection. A slight excess of fluid (arrows) is visible in the inter-cellular spaces near the endolymphatic surface. *B* basal cell;

C capillary; *IC* intermediate cell, *M* marginal cell, marker = 2 μ m.

verstein & Yules, 1971), a number of marginal cell abnormalities have been sporadically observed, including cytoplasmic vacuoles resembling those described by Nakai (1971). The significance of these changes is still completely unknown and, at present, an attempt is accordingly being made to investigate them further.

DISCUSSION

Ethacrynic acid, it is now realised, produces a wide range of metabolic effects at the molecular level and it is clearly beyond the scope of this paper to review its action in any detail. However these effects can be broadly divided into two main groups, namely the specific inhibition of a number of enzyme systems (including Na⁺ K⁺ activated ATPases) and a series

of ill-understood abnormalities of membrane permeability.

The changes resulting from both types of activity are evident in the endocochlear potential recordings, where the initial rapid decline in the potential is almost certainly the result of enzyme inactivation. The magnitudes of the resulting negative potentials, it will be noted, are of precisely the same order as the negative endolymphatic potentials arising in anoxia and after ouabain administration. In consequence not only would the inhibition appear to be complete in the early stages, but there also seems to be little alteration in the permeability of the cochlear membranes at this time, for investigation has confirmed that these other negative potentials are potassium diffusion potentials and so dependent on the



Fig. 4 Adjacent area to that shown in Fig. 3 at a higher magnification. Marker = 2 μ m.

presence of the requisite normal physical characteristics (Johnstone 1965 Kuipers & Bonting 1970a).

During the subsequent period, the situation is undeniably more complex and the later marked diminution in the rate of potential recovery is most likely to be due to an increase in permeability at that stage, of at least some portion of the membranes bounding the cochlear duct, with a concomitant decrease in their electrical resistance. In this circumstance, the enzymic processes are unable to maintain the early high rate of potential increase and recovery is thus severely impeded.

In addition, it seems reasonable to expect these permeability changes to be associated with alterations in the endolymphatic composition due to increased diffusion of sodium

into and potassium out of the scala media and this is, in fact, confirmed by the analytical findings. However the ionic constitution remains normal during the initial abolition and early recovery of the normal positive endocochlear potential and such a feature provides further evidence of the absence of any permeability abnormality during this early period, when the inactivation of the enzymes is presumably too fleeting to be accompanied by any manifest anomaly of the cationic concentrations. Moreover by 2 hours the chemical composition has returned considerably towards normal despite the persistence of the membrane effects, as judged by the level of the endocochlear potential. This feature thus provides good evidence of the efficiency of the responsible ion transporting enzyme systems at this time, for the actual values four

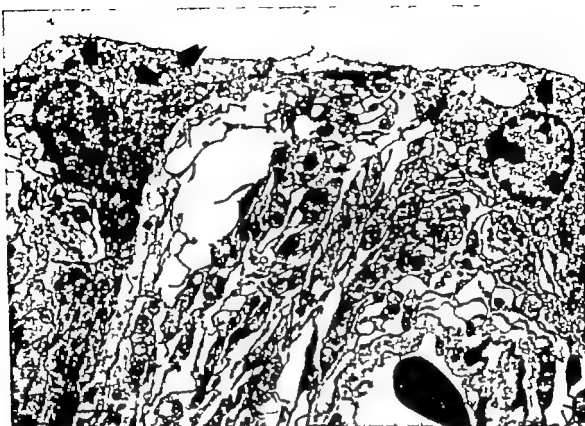


Fig. 5 The stria vascularis 2 hours post-injection in a severely affected animal. In addition to the accumulation of fluid in the inter-cellular spaces, pathological

vacuoles are present in the marginal cells (arrows). Marker = 2 μ m.

differ very considerably from those which, it can be calculated, would arise due to the physico-chemical forces alone.

The reduction in the endolymphatic chloride level and, by inference preservation of the normal bicarbonate concentration is somewhat unexpected. Whether this relative lack of change arises from preservation of the usual chloride permeability or from the functional reserve of the underlying enzyme system can only be a matter of speculation in the present state of our knowledge.

The chemical composition of the general body fluids, as revealed by the perilymph analyses, remained completely normal throughout the period under investigation. This, therefore, completely excludes the possibility that the effect of ethacrynic acid on

the endolymph and stria vascularis is secondary to some generalised electrolyte disturbance and so establishes, beyond reasonable doubt, that it must arise from the direct action of the drug upon the cochlea.

The most characteristic and consistent stria abnormality described after single injections of ethacrynic acid has been expansion of the inter-cellular spaces by accumulations of fluid (Quick & Duvall, 1970; Silverstein & Yules, 1971). In the present investigation this expansion is not associated apparently with the initial inhibition of enzyme activity and so the responsible mechanism can hardly be attributed as previously supposed, to primary depression of such activity. Instead, the appearance of the excess inter-cellular fluid coincides with the phase of altered membrane

permeability and would seem accordingly to arise directly from this effect. The preponderance of the distension, in our material, of those spaces situated between the endolymphatic surface and the capillaries adds support to this view and also suggests the excess fluid is removed, to some extent, by its passage into the capillaries.

In summary the broad outlines of the toxic action of ethacrynic acid, as far as it concerns the endolymph, can now be discerned for the first time. Initially the drug produces a short period of enzyme inactivation which is followed by a more prolonged phase of altered membrane permeability. This commences during the recovery period of the enzymic processes and is associated with the accumulation of fluid in the inter-cellular spaces of the stria vascularis. Nevertheless, it cannot be denied that a great deal of obscurity remains regarding the precise details and complex interaction of the effects produced and a more comprehensive investigation of these fundamental questions is consequently now being undertaken.

Apart from any specific considerations, however one important matter of general interest remains to be discussed. In their extremely valuable study of the action of ouabain upon the cochlear potentials, Kuipers & Bonting (1970 a) were able to show that this inhibitor produces a progressive decrease in the endocochlear potential until finally a negative potential, identical in character to the anoxic endolymphatic potential, is attained. The marked similarity between these findings and those resulting from the administration of ethacrynic acid will be immediately apparent but certain crucial differences do exist.

The changes induced by ouabain are slow in onset, largely irreversible and associated with gross ionic disturbances (Konishi & Mendelsohn, 1970). Thus secondary abnormalities, other than the very specific inhibition of the Na⁺K⁺ activated ATPase in the stria vascularis, might arguably have a role in the production of the potential alterations. Although ethacry-

nic acid is far less specific in its inhibitory activity its effects occur very rapidly and other complicating effects, as shown above, are delayed until after the initial potential changes and can therefore be excluded from consideration in this respect.

The present investigation, accordingly provides substantial support for Kuipers & Bonting's (1970 b) concept that the normal positive endocochlear potential has two components, the one a secretion potential of about +140 mV due to the activity of the stria ATPase and the other a potassium diffusion potential of approximately -40 mV arising from the selective permeability to potassium of some portion of the cochlear duct membranes. This concept is clearly of the utmost importance for our further understanding of the vital physiological functions of these membranes.

RÉSUMÉ

Le potentiel normal endocochléaire fut rapidement remplacé par un potentiel négatif du type anoxique après l'administration d'acide éthacrynique (60 mg/kg) au rat. Au début, la guérison fut rapide mais devint beaucoup plus lente après 30 min. Cependant la composition endolymphatique chimique resta sans changement jusqu'à 35 min, lorsque se produisit une augmentation progressive de la composition de sodium et une diminution de la composition de potassium, suivie après 1 heure à un retour normal graduel. Ainsi, la drogue causa probablement une inhibition momentanée des enzymes striales et ensuite des anomalies dans la perméabilité de la membrane. L'accumulation caractéristique du fluide inter-cellulaire dans la stria vascularis fut associée avec et semble survenir de ces changements de perméabilité. De plus, les conclusions supportent fortement la conception que le potentiel endocochléaire a deux composantes, un potentiel positif de sécrétion et un potentiel négatif de diffusion de potassium.

ZUSAMMENFASSUNG

Das normale endocochleäre Potential bei der Ratte wurde sehr schnell durch ein negatives Potential der Sauerstoffmangel-Art ersetzt nach Verabfolgung von Ethacrynsäure (60 mg/kg). Anfänglich erholt sie sich schnell, nach 30 Minuten wurde die Erholung aber erheblich langsamer. Die Endolymphatisch-chemische Zusammensetzung blieb jedoch 35 Minuten lang unverändert, woraufhin eine fortschreitende Zunahme in der Natrium- und Abnahme in der Kalium-

zentration erfolgte, denen eine Stunde nach Beginn, eine allmähliche Rückkehr zur Norm folgte. So scheint die Droge vorübergehende Hemmung der strahlen Enzyme zu bewirken wie auch spätere Anomalien in der Membran-Durchlässigkeit. Die charakteristische Anhäufung interzellulärer Flüssigkeit in der Stria Vascularis war mit diesen Durchlässigkeitsveränderungen verbunden und schien aus ihnen hervorzugehen. Zusätzlich unterstützen diese Befunde erheblich die Auffassung, dass das endocochleare Potential zwei Komponenten hat: Ein positives Annscheidungspotential und ein negatives Kalium Diffusionspotential.

REFERENCES

- Bosher S. K. & Warren, R. L. 1968 Observations on the electrochemistry of the cochlear endolymph of the rat: a quantitative study of its electrical potential and ionic composition as determined by means of flame spectrophotometry *Proc Roy Soc Biol* 171 227
- 1971 A study of the electrochemistry and osmotic relationships of the cochlear fluids in the neonatal rat at the time of the development of the endocochlear potential. *J Physiol* (Lond.) 212 739
- Cohn, E. S., Gordes, E. H. & Brundlow S. W. 1971 Ethacrynic acid effect on the composition of cochlear fluids. *Science* 171 910.
- Johnstone H. M. 1965. The relation between endolymph and the endocochlear potential during anoxia. *Acta Otolaryng* (Stockh.) 60 113
- Konishi, T. & Mendelsohn, M. 1970. Effect of ouabain on cochlear potentials and endolymph composition in guinea pigs. *Acta Otolaryng* (Stockh.) 69 192.
- Kalipers, W. & Bonting, S. L. 1970 a The cochlear potentials. I. The effect of ouabain on the cochlear potentials of the guinea pig. *Pflügers Arch Ges Physiol* 320 348.
- 1970 b The cochlear potentials. II. The nature of the cochlear endolymphatic resting potential. *Pflügers Arch Ges Physiol* 320 359
- Mathog, R. H., Thomas, W. G. & Hudson, W. R. 1970. Otorotoxicity of new and potent diuretics. *Arch Otolaryng* (Chic.) 92 7
- Matz, J. H., Beal, D. D. & Krames, L. 1969 Otorotoxicity of ethacrynic acid. *Arch Otolaryng* (Chic.) 90 152
- Nakai, Y. 1971. Electronmicroscopic study of the inner ear after ethacrynic acid intoxication. *Pract Otorhinolaryng* (Basel) 33 366.
- Prazma, J., Thomas, W. G., Fisher, N. D. & Preslar, J. M. J. 1972 Otorotoxicity of the ethacrynic acid. *Arch Otolaryng* (Chic.) 93 448.
- Quick, C. A. & Dravall, A. J. 1970. Early changes in the cochlear duct from ethacrynic acid. *Laryngoscope* 80 954
- Silverstein, H. & Yulek, R. B. 1971 The effect of diuretics on cochlear potentials and inner ear fluids. *Laryngoscope* 81 873
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DISCUSSION

D. Härding: Could the dilated intercellular spaces of the stria represent increased active transport of some ion (perhaps sodium) with associated water movement similar to that which has been observed by Diamond & Torrey in gall bladder epithelium?

J. Stakler: A new antibiotic substance, which was suspected to be ototoxic, has been studied in Uppsala. It is called Bicomyrin and belongs to the streptomycin group. As shown first by Japanese colleagues, Bicomyrin sometimes has remarkable effect on squamous cell carcinoma. Injected into the middle ear in experimental animals it has produced hair cell damage in the inner ear similar to that seen after streptomycin and other allied drugs. We have also observed vascular changes in the spiral ligament and stria vascularis. These changes ranged from obstruction to disappearance of the vessels. When administered subcutaneously and intraperitoneally to guinea pigs, no ototoxic effects have been observed using current morphological methods. We think that studies of the labyrinthine blood vessels must be included in any investigation of ototoxic effects, as Dr Bosher has so nicely documented.

S. K. Bosher (Reply) to Mr Härding: I think the matter you have raised is of fundamental importance and we are therefore attempting to investigate it further. The restriction of the fluid extension to those spaces situated between the endolymph and the capillaries certainly suggests this to be the pathway of the fluid movement. In addition, our findings indicate that, in all probability the fluid enters the interstices of the stria by passive diffusion because of the increased membrane permeability but we have no evidence, of course, about the mode of its removal. However, Kimura has demonstrated (in the paper by Silverstein & Yulek) that the expansion of the intercellular spaces is still present after 2-3 weeks and such a long persistence seems to me to be more in line with a passive rather than an active mechanism.

To Mr Stakler I was very interested in your comments for I believe, like you, that it is essential to investigate the effects of ototoxic agents upon all the cochlear structures, including the blood vessels, if we are to fully understand their action. However the assessment of the relative importance of these varied and inter-acting effects will clearly cause enormous difficulties.

ZUR SEKRETORISCHEN AKTIVITÄT DER INTERDENTALZELLEN DES LIMBUS SPIRALIS

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Abstract: Elektroenmikroskopische Untersuchungen am Epithel des Limbus spiralis lassen erkennen, daß in den Zisternen des endoplasmatischen Retikulums und im Golgi-Komplex ein Sekret produziert und kontinuierlich in die Endolymphe abgestoßen wird. Hier verliert sich das zu Bläschen geformte Sekret anscheinend im Fasersystem der Deckmembran. Histochemische Reaktionen zeigen, daß es sich hierbei vorwiegend um Glykoproteide und disulfidbrückenreiche Substanzen handelt, aus welchen sich dann erst im Lumen der Endolymphe die Tektorialmembran unter dem Einfluß von Natriumionen zu ihrer bekannten Gestalt formiert. Morphologische und histochemische Ähnlichkeiten mit dem Sekretionsprodukt des Subkommissuralorgans im dritten Ventrikel des Vertebratengehirns, dem sog. Reissner'schen Faden, lassen daran denken, daß ebenso wie dieser auch die Deckmembran eine ionenstabilisierende Funktion haben unter.

amphorenartig gestalteten Interdentalzellen des Limbus spiralis kontaktieren mit dem größten Teil ihrer im Limbusstützgewebe eingebetteten Zelloberfläche mit natriumreicher Perilymphe (v. Ilberg, 1968). Ihre hexagonale, endolymphatische Oberfläche zeigt zentral eine trichterförmige Vertiefung auf und ist von einer im Licht und Elektronenmikroskop amorph erscheinenden Grundsubstanz überlagert. Diese Substanz ordnet sich zu feinsten Protofibrillen und läßt sich über die Lippe des Limbus spiralis hinaus in das Lumen der Scala media als sog. Tektorialmembran verfolgen. Diese Membran gehört zu den phylogenetisch konstanten Gebilden des akustischen Organs von den niederen Vertebraten bis hinauf zu den Primaten (Schenk, 1963; Wever 1971). Trotz zahlreicher Untersuchungen morphologischer, histochemischer

und biochemischer Art ist über ihre Entstehung und Funktion außer einigen Theorien bis heute wenig bekannt geworden. Wegen ihrer engen topographischen Beziehung zu den Interdentalzellen des Limbus spiralis, zum Epithel des Sulcus internus und zu den Sinuszellen des Corti'schen Organes wurde für jede dieser drei Zellarten bereits eine Mitbeteiligung am Aufbau der Deckmembran diskutiert. Die Arbeiten von Bélanger (1953), Dohleman & Ormerod (1960), Iurato (1962), Voldrich (1967) v. Ilberg (1968), Lim (1969, 1970), Ishiyama et al. (1970) haben gezeigt, daß die Interdentalzellen an ihrer endolymphatischen Oberfläche Bläschen in das Lumen der Scala media absondern, deren Inhalt möglicherweise dem Aufbau der Tektorialmembran dient. Über die chemischen und physikalischen Eigenschaften dieser Stoffe gibt es bislang keine Aussagen. Zwar weiß man, daß die Tektorialmembran neutrale Mucopolysaccharide in großer Menge und zu einem geringeren Anteil auch saure Mucopolysaccharide enthält, zudem verschiedene schwachere Verbindungen (Schäffle, 1971); ein direkter Nachweis ähnlicher topochemischer Verbindungen im Cytoplasma der Interdentalzellen steht jedoch noch aus.

METHODIK

Zur Untersuchung dienten Meeresschweinchen (200-400 g Körpergewicht). Für die elektronenmikroskopische Untersuchung erwies sich die

korporale Perfusion mit dem Fixierungsgemisch nach Bouin oder Allen B. 15 nachfolgender Immersionsfixation und anschließender Entkalkung in EDTA als besonders günstig. Die Paraffinschnitte wurden mit folgenden Methoden angefärbt. Aldehydthioninreaktion nach Paget (1959) Chromalaunhaematoxilin-Phloxin nach Gomori-Bargmann (Pearse, 1960) Paraldehydfuchsin nach Gabe (1956) Alcanblaufärbung nach vorhergehender Oxydation nach Adams & Sloper (1956). Die Reaktionen wurden nach der Vorschrift von Romels (1968) ausgeführt. Für die elektronenmikroskopische Untersuchung fixierten wir mit 2,5%igem Glutaraldehyd (Karlsson & Schultz, 1965) oder 6,4%igem Glutaraldehyd (Rewcastle, 1965) durch korporale Perfusion und anschließende Immersionsfixation zwei Stunden lang. Die Präparate wurden in 1%igem Osmiumtetroxyd (Palade, 1952) fixiert. Die Einbettung erfolgte in Epon 12 (Luft, 1961) oder im Spurr'schen Gemisch (1969). Soweit keine histochemischen Reaktionen ausgeführt wurden, erfolgte Nachkontrastierung mit Bleichtrat und Uranylazetat. Lokalisation der Natriumionen benutzten wir die modifizierte Methode nach Kornick (1963) Fixiert wurde in einem Teil 2%igem Thioanthonat und einem Teil 4%igem Osmiumtetroxyd bei einem pH von 7,6. Zum elektronenmikroskopischen Nachweis von Natrium Mucoproteiden gingen wir nach der Vorschrift von Rambourg (1967) und Rambourg et al. (1969) vor. Zum Nachweis der Transportfunktion zytopenptischer Bläschen die Interdentalzellen injizierten wir 5-10 Minuten vor der Fixation 50-100 μ l Thionindioxyd in die Cisterna cerebello-medullaris. Zur Überprüfung möglicher Mucoproteinstränge in der Endolymphflüssigkeit entnahmen wir dem lebenden Tier Endolymph und setzten gleiche Mengen von 2%iger Phosphorsäure in 1%igem Ammoniumazetatpuffer bei einem pH von 7,0 zu. Mit einer Mikrokapillare wurde ein Tröpfchen dieses Gemisches auf ein Kupfernetz gegeben, die Flüssigkeit unter Stickstoffatmosphäre ange trocknet und dann im Elektronenmikroskop untersucht.

BEFUNDE

Nach Fixation im Bouin'schen Gemisch und Entkalkung mit EDTA lassen sich mit der PAS-Färbung nach Hotchkiss und Magnus unterschiedlich große Sekretvakuolen supranukleär im Grundzytoplasma der Interdentalzellen darstellen. In einem Teil der Vakuolen findet man stark PAS-positive kugeförmige Einschlüsse, deren Übertritt in die ebenfalls sehr stark PAS-positiv reagierende Tektorialmembran in der Regel zu verfolgen ist. Mit Chromalaunhaematoxilin-Phloxin und Aldehydthionin gelingt es, feine, tief blaue Granula im Grundzytoplasma der Interdentalzellen nachzuweisen. Mit diesen hauptsächlich auf Disulfidbrücken (Neurosekret) zielenden Färbemethoden zeigt die Tektorialmembran ebenfalls eine stark positive Reaktion. Nach Glutaraldehydfixation und nachfolgender Osmierung gelingt es allerdings in den Semidick schnitten nicht, die oben erwähnten wasser klaren Sekretvakuolen darzustellen. Dagegen färben sich auch hier mit PAS, Chromalaunhaematoxilin und Aldehydthionin neben der Tektorialmembran zahlreiche Granula im Grundzytoplasma der Interdentalzellen an. Bei der Überprüfung der Natriumverteilung im Bereich des Limbus spiralis und den übrigen Strukturen der Scala media mit der modifizierten Methode nach Kornick ist die Tektorialmembran das einzige Gebilde, welches in auffälliger Weise Natriumpyranthionatniederschläge anreichert. Insbesondere ist festzustellen, daß eben dort, wo im Bereich der ersten Interdentalzellen vor dem Ansatz der Reissner'schen Membran die Grundsubstanz der Tektorialmembran erstmalig licht mikroskopisch auf der Oberfläche sichtbar wird, auch Natriumniederschläge angetroffen werden. Über den epithelalen Zellen

Wir danken Mrs T. Forte, M.D. Donner Laboratory and Donner Pavilion, Berkeley für Ihre hilfreiche Unterstützung bei der Durchführung dieses methodischen Abschnittes.



Abb. 1 (a) Semidünnschnitt durch den Limbus spiralis der rechten Scala media. Chromalaunhaematoxylin-Phloxin-Färbung. Neben der positiven Reaktion im Bereich der Tektorialmembran sind besonders im zentralen Anteil des Limbus spiralis der zu jeder einzelnen Interdentalzelle gehörige Sekretbelag (→) und die scharf gezeichneten positiv gefärbten Interzellularfugen deutlich hervorgehoben. *al* = Sulcus internus; *SM* = Scala media, *RM* = Reissner'sche Membran. Fix.

Glutaraldehyd; 500× (b) Positive Reaktion einiger Sekretgranula im Grundzytoplasma der Interdentalzellen (→) und der Tektorialmembran. Fix. Glutaraldehyd-Aldehythionia. 1400× (c) Nach Borie-Fixation ist das Cytoplasma im Gegensatz zur Fixation mit Glutaraldehyd vakuolig aufgetrieben. Intrazellulär stellen sich Kondensate eines PAS-positiven Materials im optisch leeren Grundcytoplasma dar. 800

bus, die nicht von der Tektorialmembran werden liegen auch keine Pyroantimonatpartikelchen (Abb. 1 2).

Ultrastrukturell weisen die Interdentalzellen typische morphologische Kriterien für sekretorisch aktive Zellen auf. So findet sich in der Regel supranukleär ein ausgeprägtes lamelläres System von Schläuchen des endoplasmatischen Retikulums, die einen dichten Ribosomenbesatz zeigen. An ihren Enden schnüren sich Bläschen ab, die teilweise noch mit Ribosomen besetzt sind und treten offensichtlich zu dem stets nahegelegenen Golgi-Komplex in enge Beziehung. Von dort aus lassen sie sich mit unterschiedlich dichtem Inhalt bis an die Oberfläche der Interdentalzellen verfolgen. Sie wölben die Zellmembran in Richtung Endolympe vor ein Vorgang der häufig als mikrovilliartige Zellkonfiguration im-

portiert. Diese zytoplasmatische Ausstülpungen schnüren sich ab und werden mit ihrer Hilfe in die Grundsubstanz der Tektorialmembran ausgestoßen. Sehr deutlich läßt sich im Elektronenmikroskop eine Beziehung zwischen Grundsubstanz der Tektorialmembran und intrazytoplasmatischen Bläschen erkennen. Nur dort wo im Grundzytoplasma Bläschen zur Oberfläche der Interdentalzellen wandern und in die Endolympe ausgestoßen werden ist auch die amorphe Grundsubstanz der Tektorialmembran nachweisbar. Die an weitesten lateral zum Ansatz der Reissner'schen Membran hin gelegenen Limbusepithelzellen enthalten nur ganz vereinzelt wasserklare zytoplasmatische Vesikel an ihrer Oberfläche fehlt der amorphe Belag, den man üblicherweise weiter medial auf dem Limbusepithel erkennen kann (Abb. 3).

Daneben trifft man im Grundzytoplasma der Interdentalzellen auf eine weitere Art von Zytopempsisbläschen, die sich, wie mehrfach beschrieben, aus der lateralen Zellbegrenzung in das Zellinnere einstülpen, von dort ebenfalls an die Oberfläche wandern und in die Endolympe entleert werden. Nach Injektion von Thorotrast in den Liquor cerebrospinalis enthalten jene Bläschen bereits nach wenigen Minuten Thorotrastpartikelchen, die man schließlich auch in der Tektorialmembran wiederfindet.

Neben den beschriebenen Zytopempsisbläschen heben sich Vesikel von unterschiedlicher Größe und relativ elektronendichtem Inhalt ab die über das gesamte Zytoplasma verstreut liegen. Ihre enge Beziehung zum Golgi-Komplex läßt auf eine Mitbeteiligung dieses Systems bei der Aufbereitung ihres dichten Inhaltes schließen. Gelegentlich trifft man auch relativ große zytoplasmatische Einschlüsse, deren membranbegrenzter elektronendichter Inhalt, sowie die enge Beziehung zu Lamellen des Golgi-Apparates und den dunklen Vesikeln daran denken läßt, daß es sich um eine Sekretspeicherungsform handelt. Unter Anwendung der PA-Chrom Silber methenamin-Methode nach Rambourg (1967) konnten wir in vielen zytoplasmatischen Vesikeln Glykoproteidverbindungen lokalisieren. Ebenso wie diese reagierte die Tektorialmembran stark positiv allerdings auch das Gitterfasernetz des Limbusstützgewebes (Abb. 4).

Mit der Komnick'schen Kaliumpyroantimonatmethode gelang es im elektronenmikroskopischen Bereich nicht, im Lumen der Zytopempsisbläschen einen positiven Reaktionsausfall zu erkennen. Dagegen läßt sich anhand benachbarter Zellstrukturen zeigen, daß der zytopempsische Transport von Natriumionen in anderen Epithellen der Scala media möglich ist (positiv im Bereich der Reissner'schen Membran). Auch die perilymphatische Umgebung der Interdentalzellen im Bereich der Huschke'schen Zähne enthält zahlreiche Natriumpyroantimonatniederschläge Sowohl im Lichtmikroskop als auch

im Elektronenmikroskop wird sehr deutlich, daß die Tektorialmembran einen besonders starken Reaktionsausfall beim Nachweis auf Natriumionen zeigt. Die filamentäre Struktur der Deckmembran ist durchsetzt von zahllosen Pyroantimonatniederschlägen. Im Zytoplasma der Interdentalzellen dagegen lagern ausschließlich die großen elektronendichten membranbegrenzten Sekretpakete an ihrer Oberfläche Natriumpyroantimonat an.

Schließlich untersuchten wir noch mit der „negative staining Methode“ lebend frisch entnommene Endolympe und unfixierte Tektorialmembran und fanden daß in der Endolympe zahllose fadenartige Strukturen vorhanden sind, die große Ähnlichkeit mit den Fibrillen der Tektorialmembran haben. Handelt es sich dabei tatsächlich um Glykoproteide, so wäre zu erwarten, daß diese irgendwo im Wandepithel des Ductus cochlearis resorbiert werden, falls sie nicht vorher enzymatisch aufgelöst wurden. Deshalb kontrollierten wir mit den oben beschriebenen histochemischen Methoden das Wandepithel des Endolymphaumes und fanden sowohl lichtmikroskopisch als auch elektronenmikroskopisch im Epithel des Sulcus externus und im Bereich der epithelialen Schicht der Reissner'schen Membran einen positiven Reaktionsausfall auf neutrale Glykoproteide (Abb. 5).

DISKUSSION

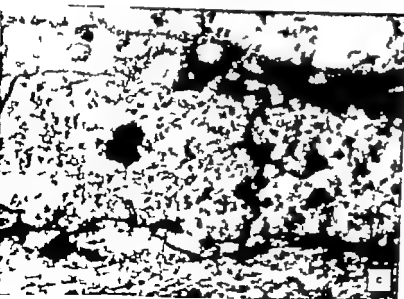
Bereits Voldrich hatte 1967 gezeigt, daß die Interdentalzellen rhythmisch ein wasserklares Sekret in die Endolympe abgeben. Eine Anfärbbarkeit des Vakuoleninhaltes oder des Restzytoplasmas mit PAS gelang jedoch nicht. Auch Iurato (1962), v. Ilberg (1968), Lim (1970), Ishiyama et al. (1970) haben aufgrund ihrer morphologischen Befunde auf eine sekretorische Funktion der Interdentalzellen geschlossen. Enzymhistochemische Untersuchungen am Limbusepithel (Vosteen, 1960; Gerhardt, 1962; Ishii & Balogh, 1966; Lim, 1970; Nomura & Balogh, 1964



Abb 2 (a) Nach Kalium-Pyrosulfat-Osmiumfixation findet man auf der perilymphatischen Oberfläche der Retikularmembran zahlreiche Niederschläge. Im Endolymphraum fallen lediglich im Bereich des Glykoproteidbelages über dem Limbus spiralis Niederschläge aus. Dort wo das Limbus epithel frei von Glykoproteidbelag ist (→) fehlen auch die Niederschläge. 500 x



(b) Die Tektorialmembran ist durchsetzt von Natrium Pyrosulfatniederschlägen. Fix. wie a. 900



(c) Ausschnitt eines apikalen Anteiles einer Interdentalzelle im elektronenmikroskopischen Bild. Die Glykoproteidschicht über den Interdentalzellen enthält zahlreiche Natriumniederschläge, während das Grundcytoplasma der Interdentalzellen und das Cytoplasma der Zellen frei von Pyrosulfatniederschlägen sind. Im Limbusgewebe dagegen sind positive Niederschläge (→) vorhanden. Fix. wie a. 6400



1. (a) Elektronenmikroskopischer Ausschnitt 1 der endolymphatischen Oberfläche zweier benachbarter Interdentalzellen. Über der einen Zelle (a) liegt ein grauer Belag mit vereinzelt optisch leeren Membranriegen (). Im darunterliegenden Cytoplasma erkennt man zahlreiche scharf begrenzte Bläschen mit fein granuliertem Inhalt, die in die Tektorialmembran abgesondert werden. Dagegen zeigt die benachbarte Interdentalzelle () nicht diese scharf

begrenzten Bläschen im Grundcytoplasma, dementsprechend fehlt auch der graue Belag auf der endolymphatischen Zelloberfläche. Fix. Osmium (Glutaraldehyd, 3 000 (b) Die Cisternen des endoplasmatischen Retikulums schließen an ihren Polen Bläschen (→) ab, welche zur endolymphatischen Zelloberfläche wandern. M = Mitochondrium K = Zellkern. Fix. wie 3 a. 32 000

1971) bestätigen anhand des Enzymmusters von oxidativen und hydrolytischen Enzymen die Annahme einer Beteiligung dieser Zellen bei einem Sekretionsprozeß.

Allerdings war es bislang noch nicht gelungen über die chemischen und physikalischen Eigenschaften des Sekretionsproduktes eine genaue Aussage zu machen. Die von uns angewandten histochemischen und histologischen Untersuchungsmethoden ergaben sowohl im Licht als auch im elektronenmikroskopischen Bereich, daß die Interdentalzellen ein Sekret produzieren, das die gleichen histochemischen Eigenschaften zeigt, wie die Tektorialmembran selbst. Neben der stark positiven Reaktion auf PAS gelang auch der Nachweis von sulfidbrückenreichen Anteilen im Grundzytoplasma der Interdentalzellen. Wir schließen daraus, daß die Tektorialmembran als Sekretionsprodukt der Interdentalzellen aufzufassen ist. Als weiteren Beweis möchten wir die Tatsache anführen, daß im elektronenmikroskopischen Bild das Limbus-epithel nur an den Stellen von Deckmembran überlagert ist, wo zytopenetische Bläschen an die Oberfläche ausgestoßen werden. Allerdings erklärt sich aus dem alleinigen Sekretionsvorgang noch nicht die konstante Konfiguration der Tektorialmembran. Es wird weiterhin diskutiert, daß Ionen die Form von Mucopolysacchariden stabilisieren (Davis, 1958; Vilstrup & Jensen, 1954; Jensen & Vilstrup 1960). Bei der Überprüfung der Natriumverteilung im Bereich des Innenohres konnten wir zeigen, daß im endolymphatischen Kompartiment die Tektorialmembran selektiv Pyroantimonatniederschläge anreichert. Da mit dieser Methode nicht nur Natrium-, sondern auch Magnesium- und Kalziumionen ausgefällt werden können (Komnick, 1963; Sumi, 1971; Sumi & Swanson 1971; Klein et al., 1972; Forte 1970 persönl. Mitteilungen) muß gefolgert werden, daß die Tektorialmembran reich an Ionen dieser drei Elemente ist. Aus den Untersuchungen von Naftalm et al. (1964) geht jedoch hervor, daß im Hydrolysat der Tektorialmembran nur



Abb. 4 Mit der PA-Chr-Silber Methenamin-Methode nach Rambourg lassen sich im Grundcytoplasma der Interdentalzellen und im Bereich der Tektorialmembran (TM) neutrale Glykoproteide elektronenmikroskopisch nachweisen. Fix. Glutaraldehyd ohne Osmium. 9750 \times Laser. Entsprechende Interdentalzelle im Lichtmikroskop nach Chromalaunhaematoxylin-Phloxin-Färbung und gleicher Fixation 1:500

sehr geringe Mengen Magnesium und Kalzium ebenso wie in der Endolymph selbst zu finden sind. Deshalb können wir zum gegenwärtigen Zeitpunkt und unter kritischer Berücksichtigung der methodischen Spielbreite vermuten, daß der Reichtum an Pyroantimonatniederschlägen in der Tektorialmembran ihren hohen Gehalt an Natriumionen widerspiegelt. Somit wäre es vorstellbar, daß die Glykoproteide der Tektorialmembran in ihrer Funktion als Polyelektrolyte Natriumionen anlagern und daß erst dadurch die Deckmembran ihre Form behält. Zum anderen ist zu bedenken, daß durch die Sekretion der Interdentalzellen laufend neue Valenzen frei werden und zur erneuten



Abb. 5. Negativ statilog mit Phosphor-oxiframsäure von frisch entnommener Tektorialmembran (5 a, 32 000 \times) und Endolympha (5 b, 10 600 \times). In der Endolympha findet man fadenartige Strukturen ähnlicher Konfiguration wie im Negativbild der Tektorialmembran.

Anlagerung von Ionen zur Verfügung stehen. Unter Berücksichtigung dieser Möglichkeiten könnte man den Glykoproteiden der Tektorialmembran auch eine Funktion als Ionenfänger zuschreiben auf diese Weise wird das Ionenmilieu in der Endolympha insbesondere im Bereich der Haarzellen konstant gehalten. Änderungen dieses Gleichgewichtes, hervorgerufen durch versiegende Sekretion der Interdentalzellen oder durch ein Über-

angebot an Natriumionen, müßten zu erheblichen osmotischen Veränderungen im Bereich der Endolympha führen. Einen morphologischen und funktionellen Beweis für unsere Vorstellungen liefern die Befunde von Voldrich (1968). Dem Autor gelang es zu zeigen, daß nach Ruptur der Reissner'schen Membran und der dadurch hervorgerufenen Durchmischung von Endo- und Perilymphe es zu einer meßbaren Aktivität im Sinne einer

Sekretionssteigerung der Interdentalzellen kommt. Ähnliches Verhalten der Lumbus-epithelzellen stellte Voldrich (1968) auch nach Beschallung seiner Versuchstiere fest. Nakashima et al. (1971) sowie Suga et al. (1970) konnten nach starker akustischer Stimulation ein Absinken der K^+ -Konzentration und ein Ansteigen des Na^+ -Gehaltes in der Endolymph von Meerschweinchen messen. Diese Ergebnisse passen sehr gut in das Konzept einer Abhängigkeit der sekretorischen Aktivität des Limbus-epithels vom Na^+ -Gehalt der Endolymph. Die erhöhte Aktivität der Interdentalzellen könnte als gegenregulatorischer Vorgang bei erhöhter Na^+ -Konzentration aufgefaßt werden und den Versuch darstellen, mit Hilfe des Sekretionsproduktes — entsprechend unseren Befunden — Na^+ -Ionen zu binden. Die von uns mit Hilfe der Negativstaining-Methode aufgezeigten fibrillären Strukturen im Bereich der Endolymph, die sehr große Ähnlichkeit mit dem Negativstaining-Bild der Tektorialmembran haben, könnten Abbauprodukte der Tektorialmembran darstellen die beladen mit Kationen vom Wandepithel des Ductus cochlearis resorbiert werden. Diese Vorstellung knüpft an die Behauptung von Dohlmans (1971 persönl. Mitteilg.) an, der den Mucopolysaccharidfäden der Endolymph von Tauben eine Mitbeteiligung bei der Osmoregulation zuschreibt.

Schließlich sei erwähnt, daß aus der vergleichenden Anatomie ein sehr ähnliches sekretorisches Organ bekannt ist. Es handelt sich dabei um das ependymale Subkommissuralorgan am Dach des dritten Ventrikels aller Vertebratengehirne, das als Produktionsstätte für den sog. Reissner'schen Faden gilt. Dieser Faden stellt einen neutralen Glykoproteidkomplex dar der Zystein, Thyrosin, Tryptophan und weitere biogene Amine sowie Sialinsäure enthält (Bargmann & Schiebeler 1952, Sterba, 1969 Sterba & Wolf 1969). Er verläuft durch den dritten Ventrikel, den Aqueductus Sylvii bis zur Endampulle des Zentralkanals und ist bei allen Vertebraten einschließlich dem Menschen konstant vor-

handen. Funktionell wird auch dem Reissner'schen Faden eine Mitbeteiligung bei der Osmoregulation des Liquor cerebrospinalis (möglicherweise Ionenaustauscherfunktion, Sterba, 1969; Arnold, 1969) zugeschrieben. Der Vorgang der Sekretbereitung in den Zellen des Subkommissuralorgans ist licht- und elektronenmikroskopisch genau untersucht und zeigt in vieler Hinsicht Ähnlichkeit mit den Befunden an den Interdentalzellen des Limbus spiralis cochleae.

RÉSUMÉ

Des recherches de microscopie électronique permettent de constater que les cellules du réticulum endoplasmique des cellules interdentaires produisent une substance qui est continuellement excrétée dans l'endolymph. Il s'agit de substances amorphes et invisibles en microscopie électronique qui sont entourées par une membrane tendre et forment des vésicules. Ces vésicules semblent se perdre entre les fibrilles de la membrane tectoriale. Des réactions histochimiques font présumer qu'il s'agit principalement de glycoprotéides neutres qui dans le milieu de l'endolymph forment la membrane tectoriale (sur l'influence du sodium). La ressemblance histochimique et morphologique de la membrane tectoriale avec l'organe subcommissural du 3ème ventricule et son produit le corde de Reissner font penser que la membrane tectoriale est un organe semblable qui possède éventuellement une fonction homéostatisatrice.

SUMMARY

Electronmicroscopically the cytoplasm of the interdental cells show typical features of secreting cells. An amorphous and less electron-dense substance is produced by the endoplasmic reticulum, packed by the Golgi-complex into vesicles of different sizes and are delivered into the endolymphatic space. There they get lost among the fibres of the tectorial membrane. Histochemical reactions allow the conclusion that neutral glycoproteids are the main components of the phocytotic vesicles as of the tectorial membrane. We assume that in the medium of the endolymph and under the influence of sodium ions the glycoproteid are stabilized and receive the wellknown form of the tectorial membrane. Morphologically and histochemically this kind of secretion of glycoproteids into a body fluid is very similar to the data well known from the so-called subcommissural organ of the 3rd ventricle in vertebrates. The subcommissural organ produces neutral glycoproteids into the CSF which form the Reissner's fibre. Both the tectorial membrane and Reissner's fibre are discussed as having ionic stabilizing properties.

LITERATUR

- Arnold, S. 1969 Beobachtungen am Subkommissuralorgan und Reissner'schem Faden der Schildkröte unter osmotischer Belastung. *Z Zellforsch* 101 152.
- Bargmann, W. & Schiebler T. H. 1952. Histologische und zytochemische Untersuchungen am Subkommissuralorgan von Säugern. *Z Zellforsch* 37 583.
- Bélauger L. F. 1953 Autoradiographic detection of 23 in the membranes of the inner ear of the rat. *Science* 118 520.
- Devle, H. 1958 Mechano-electrical theory of cochlear action. *Ann Otol* 67 789.
- Dohlman, G. F. 1971 The attachment of the cupulae, otolith and tectorial membranes of the sensory cell areas. *Acta Otolaryng* (Stockh.) 71 89.
- Dohlman, G. F. & Ormerod, F. C. 1960. The secretion and absorption of endolymph. *Acta Otolaryng* (Stockh.) 51 435.
- Ermisch, A., Sterba, G. Hartmann, G. & Freyer K. 1958. Autoradiographische Untersuchungen über das Wachstum des Reissner'schen Fadens von *Cyprinus Carpio* (L.). *Z Zellforsch* 91 220.
- Forie, T. M. & Forie, J. 1970. Histochemical staining and characterization of glycoproteins in acid-secreting cells of frog stomach. *J Cell Biol* 47 437.
- Gierhart, H. J. 1962. Zur Verteilung des DPN und TPN-Diaphorase in der Meeresschnecke. *Arch Otor Nas Kehlkopfheilk* 181 16.
- Huber, Ch. 1968. Elektronenmikroskopische Untersuchungen über Diffusion und Adsorption von Thioindoloxyl an der Membran einschnecke. *Arch Klin Exp Otor Nas Kehlkopfheilk* 192 163.
- Iseli, T. & Balogh, K. 1966. Acid phosphatase activity in the inner ear. *Acta Otolaryng* (Stockh.) 67 185.
- Ishiyama, E., Weibel, E., Knebel, E. B. Richardson, T. L. 1970. Ultrastructure of the interdental cells in mammals. *Acta Otolaryng* Belg 52 312.
- Imoto, S. 1962. Submicroscopic structure of the membranous labyrinth. III. The supporting structure of Corti's organ (basilar membrane, limbus spiralis and spiral ligament). *Z Zellforsch* 56, 40.
- Klein, T. Yen, C. & Thurston-Klein, A. 1972. Critique on the K-pyronin-staining method for semi-quantitative estimation of cations in conjunction with electron microscopy. *J Histochem Cytochem* 20 65.
- Koenig, H. & Koenig, U. 1963. Elektronenmikroskopische Untersuchungen zur funktionellen Morphologie des Ionentransportes in der Saldrüse von *Larus argentatus*. *Z Zellforsch* 60 163.
- Lin, D. J. 1969 Three-dimensional observation of the inner ear with the scanning electron microscope. *Acta Otolaryng* (Stockh.) 225 1.
1970. Morphology and function of the interdental cells. An ultrastructural observation. *J Laryng* 84 1241.
- Mara, E. 1971 The function of the interdental cells of the limbus spiralis. *Pract Otorhinolaryng* (Basel) 33 252.
- Naftalin, L., Harrison, M., & Stephens, A. 1964. The character of the tectorial membrane. *J Laryng* 78 1061.
- Nakashima, T., Meiring N. & Snow J. 1971 Cations in the endolymph with noise-induced deafness. *Arch Otolaryng* (Chic.) 94 109.
- Nomura, Y. & Balogh, K. 1964. Localization of DPNH and TPNH diaphorase activity in the cochlea by various histochemical techniques. *Laryngoscope* 74 1351.
- Rambourg, A. 1967 An improved silver methenamine technique for the detection of periodic acid-reactive complex carbohydrates with the electron microscope. *J Histochem Cytochem* 15 409.
- Rambourg, A., Hernandez, W., & Lebiond, C. P. 1969 Detection of complex carbohydrates in the Golgi apparatus of rat cells. *J Cell Biol* 40 395.
- Romels, B. 1968. *Mikroskopische Technik*. Oldenburg-Verlag, München.
- Schiltz W. 1971. *Histochemie des Innenohres*. Urban-Schwarzenberg-Verlag, München-Berlin-Wien.
- Schenk, J. 1963. *Entwicklung, Struktur und Lage der Membrane tectoria des Cortischen Organs* (eine literarische Zusammenstellung). Inaugural-Dissertation, Würzburg.
- Spurr A. 1969 A low-viscosity epoxy resin embedding medium for electron microscopy. *J Ultrastruct Res* 26 31.
- Suga, F., Nakashima, T. & Snow J. B. 1970. Sodium and potassium ions in endolymph. *Arch Otolaryng* (Chic.) 91 37.
- Sterba, G. 1969 Morphologie und Funktion des Subkommissuralorgans. In: *Zirkonventrikuläre Organe und Liquor* 8. 17 VEB Gustav Fischer Verlag, Jena.
- Sterba, G. & Wolf G. 1969 Vorkommen und Funktion der Salzlure im Reissner'schen Faden. *Histochemie* 17 57.
- Viktrup Th. & Jensen, C. E. 1954. Three reports of the chemical composition of the fluids of the labyrinth. *Ann Otol* 63 151.
- Voldrich, L. 1967 Morphology and function of the epithelium of the limbus spiralis cochleae. *Acta Otolaryng* (Stockh.) 63 503.
- 1968. Über die Epithelsekretion des Limbus spiralis in den Ductus cochleae. *Wiss Z Univ Halle* XX'71 M.H. 1 S. 118.
- Vosteen, K. H. 1960. The histochemistry of the enzymes of oxygen metabolism in the inner ear. *Laryngoscope* 70 351.
- Wever E. I. 1971 The mechanics of hair-cell stimulation. *Ann Otol* 80 786.
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DISCUSSION

H. Spoendlin: It would be interesting to know whether this polysaccharide secretion of the surface

of the interdental cells takes place during the entire adult life at similar rates or whether it is mainly an important process during the development of the tentorial membrane. Concerning the histochemical method of Koenig for the demonstration of Na, I do not think that the concentration of the reaction product at certain surfaces is significant evidence of high Na-concentrations at these sides during life. Secondary adsorption of histochemical reaction products at any surface is a common phenomenon.

I. Friedmann: Some of my questions have been touched upon by Drs Engström and Spoendlin. We have studied the spiral limbus of the embryonic mouse and have noted filaments passing from the cells into the membrane of Reissner's Laminated bodies have been described in the subcommissural organ by Wetstein et al. Similar cellular inclusions were described by Friedmann et al. in the human macula of the utricle of patients operated on for Ménière's syndrome. Do these inclusions indicate some metabolic disorder and have they been observed in the limbus cells?

H. Arnold (Antwort) zu Prof. Spoendlin:

a) Die rege Zellaktivität, erkennbar an der Formation des endoplasmatischen Retikulums, der Golgi-Komplexes, sowie anderer Zellorganellen sprechen für eine aktive Leistung der IDZ. Unsere Ergebnisse zeigen, dass die Zellen am Aufbau der TM maßgeblich beteiligt sind. Es ist anzunehmen (wir untersuchen nur ausgewachsene Tiere) daß die Sekretion das ganze Leben hindurch anhält. Über den Stoffwechsel der TM und ihren Abbau weiß man allerdings noch nichts genaues.

b) Die histochemische Methode erlaubt nur eine bedingte Aussage. Deshalb sind Kontrollversuche, Nachbestimmung etc., hier sehr wichtig. Es zeigte sich nach Oubalm-Verhandlung der Tiere signifikante Änderungen der Niederschlagsdichte im Endolymphraum und im Bereich der Reissner'schen Membran. Insbesondere ist hier der Sekus internus, Tectorialmembran, Haarzelloberfläche und Oberfläche der Stria vascularis von einem dichten Na-Pyranthionatbelag überlagert, was ohne Oubalmbehandlung nicht erkennbar wird. Dennoch erfordert diese Methode noch einer genauen Überprüfung.

An Prof. Friedmann:

a) Wir untersuchten nur ausgewachsene Mäuse von 200-400 g Körpergewicht. Fibrilläre Strukturen, die unmittelbar von der Zelloberfläche der Interdentalzellen abgehen (wie bei der Maus) fanden wir nicht.

b) Im Subcommissuralorgan liegen um die Gefäßkapillaren an der Basis der SCO-Zellen dichte Mucopolysaccharidkomplexe, die sog. periodisch strukturierten Körper Wetstein-Schink, Sanku u.a. basieren darauf hingewiesen und vermutet, daß es sich hierbei um eine besondere Form der Blut-Hirn-Schranke handelt. Im Bereich des Limbusstritzgewebes findet man solche Strukturen jedoch nicht. Ähnliche Gebilde liegen in der Regel zwischen den Böschchen, allerdings sind dort wiederum keine Kapillaren.

c) Menschliches Material haben wir nicht untersucht.

GOLGI STAINS ON THE GUINEA PIG ORGAN OF CORTI

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tract. The Golgi stain has been used to study
g of cochlear and efferent nerve fibers within
organ of Corti of the guinea pig. Impregnations
the fibers of the olivo-cochlear tract was obtained
often than of the cochlear nerve fibers. Some
nerve fibers showed branching limited to
the terminal end of their spiral traverse.

A large number of studies on the innervation
the organ of Corti were made between 1880
nd 1940 by a number of highly competent

Among these Cajal (1960), Held
(1926), Retzius (1884) and Lorente de N6
(1937) were outstanding. They made their stu-
by means of either the silver stains or the
Golgi method.

It was the consensus from these studies
that separate neurons supplied inner and
hair cells. Each inner hair cell was
nplied by several short radial nerve fibers,
as each outer hair cell received termi-
from a number of spiral fibers. The ex-
l spirals (to the outer hair cells) supplied
hair cells over a larger area of basilar
than the short radial fibers (to the
inner hair cells). Lorente de N6 (1937) con-
cluded from his Golgi studies on the rat that
spiral fibers often traversed a distance of
-third of a coil, and gave off many termi-
nals to outer hair cells along their course.

In the past 30 years, other features of innervation have been added. Rasmussen (1946) described the olivo-cochlear bundle which sends its axons into the organ of Corti. Through the efforts of a number of investigators (Ki-

mura & Versäill, 1962, Iurato 1962, Smith & Rasmussen, 1963 Iurato et al., 1968 Smith, in press) some aspects of the course of the bundle and its hair cells are now well-known. By the use of electron microscopy coupled with experimental nerve degeneration, and by means of the acetylcholinesterase stain we have been able to trace the nerve fiber bundles and to distinguish the nerve endings. Yet we know almost nothing about the course of single efferent axons, their length, or the number of hair cells they supply.

Electron microscopy has enabled us to visualize single nerve fibers, and the nerve endings on the hair cells. We (Smith & Sjöstrand, 1961) have counted the number of nerve endings on outer hair cells in several locations in the guinea pig organ of Corti. The outer hair cells in the basal coil have fewer cochlear nerve endings than those in the apical coils. Thirty small cochlear nerve endings were counted on a single cell from the third coil. Spoendlin (1966) has found there are about 20 radial nerve fibers terminating on the inner hair cells in the cat cochlea. We (Smith & Takasaka, 1971) have found there are 6 to 10 nerve endings on the inner hair cells of the chinchilla. But, before the present study the best anatomical information available about branching of the external spirals came from Lorente de N6 (1937). Recent neurophysiological studies from Kiang (1965) and others have raised questions about the validity of previous reports because response curves from single nerve fibers are more

This work was supported by N.I.H. grant N.S. 08813

sharply tuned than would be assumed for fibers innervating hair cells distributed over a long length of basilar membrane.

Thus lack of information is due to several morphological features. the nerve fibers are small (0.1-0.7 microns in diameter) they are unmyelinated they course together tightly packed in bundles composed sometimes of over a hundred fibers (Smith, 1967). It is not possible to trace single fibers if the silver stains are used, because all the fibers are impregnated. It is not practical to follow the fibers by electron microscopy because their pathways are long and devious.

We have used the Golgi stain as one solution to the problem. The Golgi stain has the advantage that only a few nerve fibers in a preparation are impregnated. It is thus sometimes possible to trace the course and ramifications of a single nerve fiber for some distance. We have been able to do this for the guinea pig cochlea. The material presented here is preliminary in nature as the studies are not yet complete. It will be necessary to study many more nerve fibers in different segments of the basilar membrane before any conclusions can be made.

MATERIAL AND METHODS

Twenty-one guinea pigs (42 ears) were used in the study. Nineteen were young adults and 2 were newborn animals. All were treated in a similar but not identical manner. The rapid Golgi method as described by Valverde (1970) was used. Variations were made in the timing sequence and in the manner in which the organ of Corti was exposed to the impregnating solutions. Most of the ears were prepared as follows:

The animals were anesthetized by Dial with urethane. After decapitation the temporal bones were quickly removed and fixed by intralabyrinthine perfusion with chromeosmium. This is a 0.2% solution of osmium tetroxide in 2.4% potassium dichromate. The ears were impregnated in this for 7 days. This

was followed by a 0.75% silver nitrate solution. Our usual schedule was, 7 days in chromeosmium, one day in silver nitrate, 6 days in chrome-osmium, 3 days in silver nitrate, 5 days in chrome-osmium and finally 3 days in silver nitrate. The ears were then dehydrated in ethanol. The entire organ of Corti of each cochlea was then dissected free from the modiolus, and, after completion of the dehydration, mounted on slides to be examined for nerve staining.

RESULTS

One of the problems that we encountered was the presence of heavy precipitate which interfered with visualization of the stained nerve fibers. A second problem, inherent in Golgi stains, was the unpredictability of the stain. If scala tympani was not opened widely enough to permit a ready diffusion of the impregnating solutions, there was no nerve staining. If the opening was too large, the precipitate could be so great as to obscure the stained nerve fibers. There was no staining at all in 7 ears. Too many nerve fibers were stained to permit analysis of innervation in other ears. The third problem arose from the fragility of the specimens. The prolonged impregnation and soaking in the chrome-osmium and silver nitrate produced a marked brittleness of the tissue. The nerve fibers were easily broken during the dissection dehydration and mounting procedures. Embedding in one of the epoxies may be a solution to this problem. We are at present investigating this.

Nevertheless, despite these drawbacks, we have obtained some interesting information. Both the cochlear nerve dendrites and the afferent axons were stained. Only a few fibers will be described in this preliminary report.

Efferent axons and terminals

The axons and terminals of the olivo-cochlear tract were readily recognizable because of the size of the terminals and their location. The inner spiral and the tunnel bundles were readily

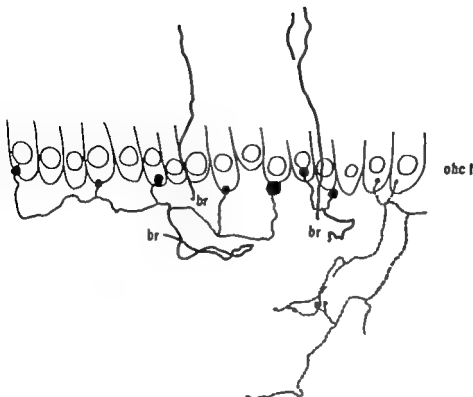


Fig. 1 Camera lucida drawing showing two efferent axons crossing the tunnel and terminating on hair cells in the first row (ohc 1). The fibers were broken (br) in preparing the specimen. The fibers at right

(dashed lines) are cochlear nerve fibers, and the terminations of the fiber illustrated in Fig. 5. (Guinea pig no. 368 L.) $\times 275$

y distinguished. These latter bundles will not be described at this time.

In many of the ears, large nerve fibers crossed the tunnel and ramified below the first row of hair cells. When observed as flat dissected specimens, mounted on a slide, they had a "grape-arbor" appearance (Fig. 1). The axon usually bifurcated with one primary branch coursing in an apical direction, the other toward the base. Single secondary branches were given off beneath the outer hair cells in the first row which terminated as large rounded bulbs on the hair cell bases. The terminal on each hair cell was usually single; occasionally double nerve endings were seen.

Fig. 1 illustrates 2 such fibers in the third cochlear coil. One axon has only 2 terminals. An adjacent axon has 5 terminals. Both axons supplied only hair cells in the first row

The impregnated end bulbs were approximately 2.5 microns in diameter.

Another efferent fiber was studied in the fourth coil from another guinea pig. Its terminal ramification covered a much larger segment of basilar membrane (0.34 mm). It had nerve endings on 34 outer hair cells in the first row; some smaller branches terminated in smaller endings on 6-8 hair cells in the second row. There was a network extending over a few hair cells which could not be traced, so that the precise number of terminals could not be determined. There may have been one or two more. Analysis of other axons in the lower part of the second coil from another ear provided additional evidence that some efferent axons supply more than one row of hair cells.

One axon in the upper part of the first coil

sharply tuned than would be assumed for fibers innervating hair cells distributed over a long length of basilar membrane.

This lack of information is due to several morphological features. the nerve fibers are small (0.1-0.7 microns in diameter) they are unmyelinated, they course together tightly packed in bundles composed sometimes of over a hundred fibers (Smith, 1967). It is not possible to trace single fibers if the silver stains are used, because all the fibers are impregnated. It is not practical to follow the fibers by electron microscopy because their pathways are long and devious.

We have used the Golgi stain as one solution to the problem. The Golgi stain has the advantage that only a few nerve fibers in a preparation are impregnated. It is thus sometimes possible to trace the course and ramifications of a single nerve fiber for some distance. We have been able to do this for the guinea pig cochlea. The material presented here is preliminary in nature, as the studies are not yet complete. It will be necessary to study many more nerve fibers in different segments of the basilar membrane before any conclusions can be made.

MATERIAL AND METHODS

Twenty-one guinea pigs (42 ears) were used in the study. Nineteen were young adults and 2 were newborn animals. All were treated in a similar but not identical manner. The rapid Golgi method as described by Valverde (1970) was used. Variations were made in the timing sequence, and in the manner in which the organ of Corti was exposed to the impregnating solutions. Most of the ears were prepared as follows:

The animals were anesthetized by Dial with urethane. After decapitation the temporal bones were quickly removed and fixed by intralabyrinthine perfusion with chrome-osmium. This is a 0.2% solution of osmium tetroxide in 2.4% potassium dichromate. The ears were impregnated in this for 7 days. This

was followed by a 0.75% silver nitrate solution. Our usual schedule was: 7 days in chrome-osmium, one day in silver nitrate, 6 days in chrome-osmium, 2 days in silver nitrate, 5 days in chrome-osmium and finally 3 days in silver nitrate. The ears were then dehydrated in ethanol. The entire organ of Corti of each cochlea was then dissected free from the modiolus, and, after completion of the dehydration, mounted on slides to be examined for nerve staining.

RESULTS

One of the problems that we encountered was the presence of heavy precipitate which interfered with visualization of the stained nerve fibers. A second problem, inherent in Golgi stains, was the unpredictability of the stain. If scala tympani was not opened widely enough to permit a ready diffusion of the impregnating solutions, there was no nerve staining. If the opening was too large the precipitate could be so great as to obscure the stained nerve fibers. There was no staining at all in 7 ears. Too many nerve fibers were stained to permit analysis of innervation in other ears. The third problem arose from the fragility of the specimens. The prolonged impregnation and soaking in the chrome-osmium and silver nitrate produced a marked brittleness of the tissue. The nerve fibers were easily broken during the dissection, dehydration and mounting procedures. Embedding in one of the epoxies may be a solution to this problem. We are at present investigating this.

Nevertheless, despite these drawbacks, we have obtained some interesting information. Both the cochlear nerve dendrites and the efferent axons were stained. Only a few fibers will be described in this preliminary report.

Efferent axons and terminals

The axons and terminals of the olivo-cochlear tract were readily recognizable because of the size of the terminals and their location. The inner spiral and the tunnel bundles were readily

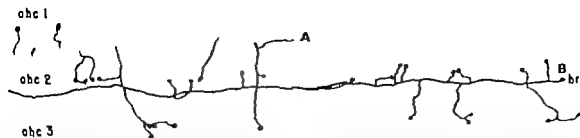


Fig. 4. Camera lucida drawing of terminal ends of two provisional cochlear nerve dendrites from the lower coil. A, fiber terminates on hair cells in the

first (ohc 1), second (ohc 2) and third rows (ohc 3). "B" terminates only on the second and third rows. (Guinea pig no. 395 R.) 245

second coil. "A" was 0.225 mm in length. thirteen nerve endings could be traced at its left end (precipitate was present, which have obscured others). These were traced as follows: 3 to the hair cells of the first row, 5 to the second row and 4 or 5 to the third row. "B" is another fiber which is 0.36 mm in length with branching on 0.135 of one end. There were 21-22 nerve endings on hair cells in the second row and on hair cells in the third row. This nerve fiber had no nerve endings on the hair cells of the first row. Two other nerve fibers are traced in Figs. 5 and 6. They are from different animals, but both are from the third cochlear coil. The nerve fiber in Fig. 5 is adjacent to the efferent nerve fiber in Fig. 1. It had 18 or 19 terminals on 12 consecutive hair cells in the first row and a fewer number

on a few hair cells in the second and third row. It could not be determined where the preterminals at the right ended.

The nerve fiber in Fig. 6 is from the third coil of another animal. It had 23 nerve endings on hair cells in the first row, 8 endings on the second row and perhaps 8 endings on the third row. The fiber is broken at the right, so that it may have had more nerve endings.

One or two cochleas in which the staining was very dense showed impregnation of numerous nerve fibers within the spiral lamina. These may have been the short radial nerve fibers to the inner hair cells. However, the stain and the precipitate were too great in these specimens for analysis. The cochlear nerve dendrites to the inner hair cells were not impregnated in any other specimens.

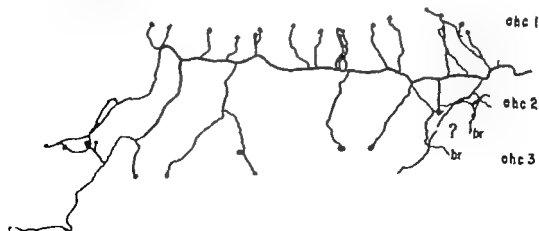


Fig. 5. Camera lucida drawing of terminal end of a cochlear nerve fiber from the lower third coil. (Guinea pig no. 368 L.) 280.

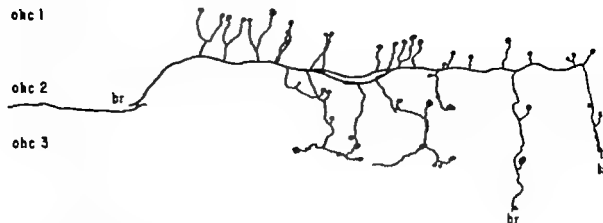


Fig 6. Camera lucida drawing of terminal end of a "provisional cochlear" nerve fiber from the third coil. (Guinea pig no. 389 L.) $\times 250$.

DISCUSSION

It should be emphasized that these are preliminary observations, based on examination of only a few of the 24 000 nerve fibers in the guinea pig cochlea (Gacek & Rasmussen, 1961). Further studies are necessary in order to come to valid conclusions. For example there seems to be an important difference between the cochlear nerve fibers seen in the lower basal coil, and the "provisional cochlear" fiber found in the upper part of the cochlea. Each of the 3 fibers found in the lower basal coil innervated only a single row of hair cells, whereas the apical nerve fibers supplied more than one row and often hair cells in all 3 rows. Yet our information is based on 3 nerve fibers stained in the first coil of only one ear and we are not really certain the apical nerve fibers are cochlear nerve dendrites.

Several reasons for the provisional identification of the fibers in the second, third and fourth coils as cochlear have been given. The fact that all the terminals, whether on hair cells from first, second or third rows are of the same diameter seems important, especially if larger efferent terminals have been impregnated in the same specimen. If the fibers were olivo-cochlear axons, one might expect the terminals of the first row of hair cells to be larger than those on the second and

third rows in the third and fourth coils. On the other hand, some efferent axons may only have large terminals, whereas the terminals on others may be smaller. Further the accumulation of globular concentrates of the chrome-osmium seems to be one of the most common artifacts of the Golgi stain. One cannot place too much credence on the size of the end-bulbs. Nevertheless, the comparative measurements of the large and small terminals correspond to the size of efferent and cochlear nerve endings as seen in electron micrographs (Smith & Sjöstrand, 1961).

A more important criterion for the identification of the nerve fibers as cochlear is found in their mode of branching. As far as can be determined the parent nerve fibers take a spiral course beneath the hair cells for a variable distance before ramification. This is typical of what we already know about the cochlear nerve fibers from previous studies made by means of the silver stains or Golgi stain. As observed to date, the efferent axons appear to ramify immediately after they cross the tunnel. Often the parent fiber bifurcates and both branches give off secondary branches to the hair cells.

The fact that these "provisional cochlear" fibers send long branches in the neighborhood of Hensen's cells is somewhat more difficult to explain. Observations made on cochlear

after acetylcholinesterase staining have revealed that AChE-positive fibers (which are efferent) are sometimes found among the Hensen's cells (Iurato & Smith, unpub.). Obviously we need some more positive identification of these nerve fibers.

The variations in the numbers and the locations of hair cells supplied by each fiber is interesting. One efferent fiber in the apical coil supplied more than 40 hair cells, whereas just a few millimeters more basalward (in the third coil) a fiber gave branches to only 2 hair cells, and another fiber to 5. It is possible, but doubtful, that these latter represent incomplete impregnation. It is much more probable that there is a considerable variation in both cochlear and efferent nerve branching. This may be related to place along the basilar membrane. It has been shown previously that there are orderly gradations in numbers of cochlear and efferent nerve endings on the hair cells from basal to apical ends of the guinea pig cochlea (Smith & Sjöstrand, 1961). Possibly there is some reflection of this in nerve fiber ramifications.

One other feature must be emphasized particularly in relation to the efferent axons. We have made no attempts to study branching in the bony spiral lamina. Several of the efferent axons which cross the tunnel may be branches of a single parent axon of the olivo-cochlear tract which enters the modiolus. It would not be possible to trace the axons in their entire extent from the internal auditory meatus to their terminals by the methods used.

No long spiral fibers which branched along their entire course have yet been seen in the guinea pig. Lorente's (1937) findings that nerve fibers coursed for one-third of a coil giving off branches along the way should be examined in context of the size of the rat's cochlea. The basal coil in the rat is approximately $5\frac{1}{3}$ mm in length the second coil is about $3\frac{1}{2}$ mm long (Békésy 1960). One-third of a coil would be 1-2 mm. The true spiral length of one of the fibers in Fig. 2

could not be measured because it was broken, but the intact portion was 0.7 mm. The length may be comparable to those Lorente found, even though the manner of branching is not. It is possible that we simply have not yet encountered the type of fiber that Lorente described. It is also possible that this type of branching is more prevalent in the rat than in the guinea pig. Other species differences within the cochlea, such as differences in number of efferent terminals per outer hair cell in guinea pigs and chinchilla (Smith & Rasmussen, 1965) and absence of a tunnel bundle in the rat (Iurato, 1961) have already been described. Another species variation would not be surprising.

RESUME

La méthode de Golgi est employée afin d'étudier des ramifications des nerfs cochléaires et éfferentes dans l'organe de Corti du cobaye. L'impregnation des fibres olivo-cochléaires est obtenue plus fréquemment que celle des fibres cochléaires. Quelques fibres cochléaires ont démontré des branches seulement à leurs extrémités.

ZUSAMMENFASSUNG

Die Verzweigungen der afferenten und efferenten Fasern im Cortischen Organ von Meerschweinchen werden mit Hilfe der Golgi Methode untersucht. Fasern der olivo-cochlearen Bündel liessen sich leichter färbten als Cochlearis-Fasern. Die Cochlearis-Fasern zeigen in der Regel erst in den Endabschnitten ihre Verzweigungen.

REFERENCES

- Békésy, G. v. 1960. *Experiments in hearing*. McGraw-Hill, N.Y.
- Cajal, R. 1960. *Studies on vertebrate neurogenesis* (trans. by Lloyd Guth), C. C. Thomas.
- Gacek, R. & Rasmussen, G. 1961. Fiber analysis of the statoacoustic nerve of guinea pig, cat and monkey. *Ann. Rec.* 139-455.
- Held, H. 1926. Die cochlea der Säugetiere und der Vögel, ihre Entwicklung und ihr Bau. In *Handbuch der normalen und pathologischen Physiologie* (ed. Bethe et al.), II Band: Receptionsorgane I. Springer Berlin.
- Iurato, S. 1961. Submicroscopic structure of the membranous labyrinth. II. The epithelium of Corti's organ. *Z. Zellforsch.* 53-259.
- 1962. Efferent fibers to the sensory cells of Corti's organ. *Exp. Cell Res.* 27-162.

- Iurato, S., Smith, C., Eldredge, D. & Henderson, D. 1968. Electron microscopic observations and cochlear potentials after section of the crossed olivocochlear tract in the chinchilla. *Fourth Eur Regional Conf on Electron Microscopy* (Rome), p. 561.
- Iurato, S. & Smith, C. Unpub. data.
- Kiang, N. 1965. *Discharge pattern of single nerve fibers in the cat's auditory nerve*. MIT Res. Monogr., 35. MIT Press, Cambridge.
- Kimura, R. & Wersäll, J. 1962. Termination of the olivocochlear bundle in relation to the outer hair cells of the organ of Corti in guinea pig. *Acta Otolaryng* (Stockh.) 55: 11.
- Lorente de No, R. 1937. The sensory endings in the cochlea. *Laryngoscope* 47: 373.
- Rasmussen, G. 1946. The olivary peduncle and other fiber projections of the superior olivary complex. *J Comp Neurol* 84: 141.
- Retzius, G. 1884. *Das Gehörorgan der Wirbeltiere*. Samson & Wallén, Stockholm.
- Smith, C. 1961. Innervation pattern of the cochlea. The internal hair cell. *Ann Otol* 70: 504.
- 1967. Innervation of the organ of Corti. In *Submicroscopic structure of the inner ear* (ed. S. Iurato). Pergamon Press, New York.
- The efferent neural supply to the vertebrate ear. In *Adv. ORL*, S. Karger, Basel, in press.
- Smith, C. & Rasmussen, G. 1963. Recent observations on the olivocochlear bundle. *Ann Otol* 72: 489.
- 1965. Degeneration in the efferent nerve endings in the cochlea after axonal section. *J Cell Biol* 26: 63.
- Smith, C. & Takasaka, T. 1971. Auditory receptor organs of reptiles, birds and mammals. In *Contributions to sensory physiology* (ed. W. Neff). Academic Press, New York.
- Spoendlin, H. 1966. The organization of the cochlear receptor. *Adv. ORL*, S. Karger, Basel.
- Vaherde, F. 1970. The Golgi method. A tool for comparative structural analysis. In *Contemporary research methods in neuroanatomy* (ed. W. Nauta & S. Ebbesson). Springer Verlag, New York.

DISCUSSION

H. Engström The different impregnation techniques give beautiful results occasionally and, in good hands, often. Different authors have reported beautiful results with these techniques and we have used scanning electron microscopy for this purpose. A combination of many methods is necessary to get the full picture of how and where the nerve fibres run inside the organ of Corti. We have found the Mallet method to be very valuable especially for the demonstration of efferent fibres but the technique often demonstrates also afferent fibres in a beautiful manner.

H. Spoendlin I was very glad to see Dr Smith's interesting results on the nerve fibre connections of the outer hair cells in the guinea pig. These results are almost exactly the same as we have shown several years ago in the cat on the basis of numerical analysis of basilar and outer spiral fibres and the nerve endings at the outer hair cells. Also in the cat we find an average spiral extension of the outer spiral fibre of 0.6 mm with only terminal branching and nerve endings to about 10 outer hair cells. (Spoendlin, 1971a: Frequency analysis and periodicity detection in hearing (ed. R. Plomp & G. F. Smoorenburg, A. W. Sijhoff, Leiden, Holland). It is important that the findings in the cat have now been confirmed in the guinea pig. This allows us to consider it as a true innervation principle rather than only a species specific innervation pattern.

Mrs Smith (Reply) to Mr Engström and H. Spoendlin. The more different kinds of techniques which are used on the problem, the more quickly we reach a solution. It is important to close the gap between structure and physiology as rapidly as possible.

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WEITERER BEITRAG ZUR ÄTIOLOGIE DER PRESBYACUSIS

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Abstract: In der Voraussetzung, dass die Ursache der Altersschwerhörigkeit in einer progressiven Knochenablagerung in der Gegend des Tractus spiralis mit anschließender Atrophie der Nervenfasern liegt, haben wir auch andere Öffnungen für die Nerven an der Schädelbasis untersucht. Wir konnten feststellen, dass es mit dem Alter auch im Bereich der anderen Öffnungen zur Knochenablagerung, die zur Verengung derselben führt, kommt. Dieser Vorgang ist für einige Manifestationen in der Funktion dieser Nerven verantwortlich. Wir haben versucht, dies auch im Tierexperiment an Ratten zu bestätigen.

Vor fast 15 Jahren haben Sercer und ich die erste Arbeit über die Ätiopathogenese der Altersschwerhörigkeit veröffentlicht. Die ersten Befunde haben sich auf die Untersuchungen an mazerierten Schläfenbeinen der grossen Schädelammlung von Perović in Zagreb bezogen. Diese Sammlung umfasst 1300 Schädel vom Fetalstadium angefangen bis zum 90 Lebensjahr. An diesen Präparaten haben wir eine mit dem Alter progressiv fortschreitende Knochenablagerung am Boden des inneren Gehörganges, die zur Verengung und endlich zum Schwund der stieförmigen Öffnungen im Tractus spiralis foraminosus führt, gefunden. Die Knochenablagerung fand zuerst in dem der Basalwindung entsprechenden Teil des Tractus statt und schritt gegen die Modiolargegend fort. Die Verengung bzw der Schwund der Öffnungen im Tractus muss durch Kompression zur progressiven Atrophie der Akustikusfasern geführt haben.

Die Befunde an mazerierten Schläfenbeinen habe ich an den Schläfenbeinsserienschnitten bestätigen können. Die histologischen Unter-

suchungen habe ich an 114 Schläfenbeinen von 57 Individuen im Alter vom Neugeborenen bis zum 90 Lebensjahr an der Sammlung von Prof Lindsay im Otologischen Laboratorium der Universität in Chicago sowie an 40 Schläfenbeinen von 20 Individuen aus der Sammlung von Prof Kelemen (1969) in der Otologischen Foundation in Los Angeles und endlich an eigenen Schnittserien im Anatomischen Institut in Zagreb durchgeführt. Die Präparate waren beiderlei Geschlechts. Für die Mehrzahl der Individuen lag entweder ein Audiogramm oder die Krankengeschichte vor so dass die Befunde am Knochen mit den klinischen Befunden verglichen werden konnten.

Die Knochenapposition im Bereiche des Tractus spiralis ging einige Phasen durch. Zuerst fand man straffes fibröses Bindegewebe, das im polarisierten Licht Doppelbrechung zeigte. Dann wurde in dasselbe Osteoid abgelagert, das später verkalkte. Auch Gussen beschreibt den Verschluss der vaskulären Kanäle in der knöchernen Ohrkapsel, durch Ablagerung vom anorganischen blauen färbenden Material in das Bindegewebe um die Gefässe. Dieses Material wird später kalzifiziert. Sie hat das kalzifizierende Material mit dem fortschreitenden Alter immer häufiger gefunden. Das Material komprimierte die Gefässe. Diese Veränderungen hat sie in grösseren Mengen in der Schneckenkapsel und in der Gegend des Meatus acusticus internus gefunden. Die neugebildete verknöcherte Masse war acellulär.

Max Meyer (1923) hat das Zustandekommen der sog durchbohrenden Kanäle in der Labyrinthkapsel von menschlichen Feten und Jungkindern beschrieben. Er hat dabei an histologischen Schnitten ebenfalls das Wachstum des Knochens in die Dicke am Hauptgefäß durch Osteoidablagerung im Bindegewebe mit Osteoblasten gefunden.

Die Knochenablagerung im inneren Gehörgang spielt sich im Bereich des endostatischen Knochens ab aus dem die siebförmigen Formationen und der Modiolus entstehen. Die Verknöcherung des Modiolus erfolgt zuerst in Form einer netzförmigen Ablagerung von Kalk in das Bindegewebe, das die Nervenstränge umhüllt. Die Verknöcherung schreitet von der Basis gegen die Spitze fort.

In diesem Zusammenhang wollten wir folgendes feststellen:

I Ist diese Knochenablagerung im Tractus spiralis ein isolierter Vorgang im menschlichen Körper oder gibt es auch andere Stellen an der Schädelbasis bzw. am Knochen system überhaupt, wo es zur Verengung bzw. zum Verschluss der Nerven- oder Gefäßöffnungen im Knochen kommt?

II Warum wird die Basalwindung zuerst befallen?

III Ist die Knochenablagerung die Ursache der Folge der Atrophie und des Schwundes der Nervenfasern?

IV Wie verhält sich ein knöcherner Nervenkanal wenn der Nerv im Experiment durchtrennt wird?

Um die erste Frage zu beantworten untersuchten wir 1) andere Bereiche der Schädelbasis, wo die Nervenfasern ebenfalls durch ein System von siebförmigen Öffnungen hindurchtreten u. zw. die beiden vestibulären Areen und die Lamina cribrosa, 2) die Nerven bzw. Gefäßkanäle der Schädelbasis u. zw. den Canalis rotundus, das Foramen ovale, das Foramen stylomastoideum, den Canalis N. hypoglossi und das Foramen spinosum und 3) die Foramina intervertebralia in bezug auf die Osteophytenbildung der Wirbelkörper.

Aufgrund dieser Untersuchungen die alle

im Rahmen unseres Institutes durchgeführt wurden, konnten wir folgendes feststellen.

1) Die Knochenablagerung findet auch in den vestibulären Areen statt u. zw. intensiver in der sakulären als in der utrikulokampulären Gegend, was sowohl an makroskopischen Präparaten als auch an histologischen Schnitten bestätigt werden konnte. Diese Knochenablagerung könnte für einige Alterserscheinungen im Gleichgewichtsorgan verantwortlich sein (Presbyostasis). Kelemen hat bei einem togeborenen Hydrocephalus (ohne Anomalien des Ohres) Exostosen im inneren Gehörgang an die Äste des N. VIII für den Sacculus gefunden. Er äußert die Möglichkeit, dass dieselben Störungen in der vestibulären Funktion hervorrufen könnten. Die Öffnungen der Lamina cribrosa verengen und verschließen sich allmählich mit dem Alter in posteroanteriöser Richtung, was für die Altersstörungen des Riechsinnes verantwortlich sein könnte (Presbyosmie).

2) Pečina M. hat die Altersveränderungen des Canalis rotundus und des Foramen ovale am Keilbein durch welche der II bzw. III Trigeminusast durchtreten am grossen Material unseres Institutes untersucht. Er konnte feststellen, dass der Canalis rotundus bei den Frauen im Durchschnitt enger ist und dass dessen Durchmesser mit dem Alter in beiden Geschlechtern um etwa 0,4 mm abnimmt (von 3,37 im 11-20 Lebensjahr auf 2,92 im 80-90 Lebensjahr). Das Foramen ovale ist enger auf der rechten Seite und im weiblichen Geschlecht und verengt sich ebenfalls allmählich in beiden Geschlechtern mit dem Alter. Diese Altersverengung der beiden Öffnungen könnte für die Trigeminusneuralgie im mittleren und höheren Alter verantwortlich sein.

Večerina (1972) hat an demselben Material auch eine signifikante Verengung des Foramen stylomastoideum, des Foramen spinosum und des Canalis N. hypoglossi mit dem Alter gefunden. Das Foramen stylomastoideum ist bei den Frauen im Durchschnitt enger als bei den Männern und wird im Alter in beiden Geschlechtern progressiv enger. Der Prozess

ist im Alter etwas intensiver bei den Männern. Die Verengung wird besonders ausgeprägt im vierten Dezennium. Auch an den anderen erwähnten Öffnungen tritt die Verengung mit dem Alter ein, beim Foramen spinale im dritten und beim Canalis N. hypoglossi schon im zweiten Dezennium. Nach dem fünften Dezennium ist die Verengung des Foramen spinale intensiver bei den Frauen. Während die Verengung am Foramen stylomastoideum und Foramen spinale konzentrisch ist, wird der Canalis N. hypoglossi durch Stachelbildungen verengt. Diese Verengungen könnten für einige Störungen im Bereiche der entsprechenden Nerven und Gefäße verantwortlich sein.

3) Ähnlich wie die Stachelbildung am Canalis N. hypoglossi kommt es auch zur Osteophytenbildung an den Wirbelkörpern und zur abschliessenden Verengung im Bereiche der durchtretenden spinalen Nerven und Arterien was zu Störungen in den betreffenden Versorgungsgebieten führt.

Die Lage der Schnecke im Raum und folglich auch die Neigung derselben gegen den Meatus acusticus internus wechselt nach Alexander im Laufe der Entwicklung. Nach Alexander (1902) schliesst die Ebene des hinteren Bogenganges mit der Ebene der Schneckenbasis am Neugeborenen nach oben einen Winkel von 25° ein, am Erwachsenen hingegen beträgt der Winkel nur 9°. Am Neugeborenen steht die Schneckenspitze nach vorne und abwärts und am Erwachsenen nur nach vorne, so dass sich die Schnecke postembryonal aufzurichten scheint. Da zur selben Zeit der innere Gehörgang beim Erwachsenen länger sein Eingang enger und die Knochenwand des Gehörganges durch subperiostale lacunäre Knochenresorptionsherde und Anlagerung von Knochenlamellen (Meyer 1923) zackig und bucktig wird, bildet sich in der Mehrzahl der Fälle eine mehr oder weniger tiefe Bucht zwischen der unteren Wand des Gehörganges und der Basalwindung der Schnecke die am tiefsten unten zu liegen kommt. Die Zirkulation des Liquors müsste



Fig. 1

in dieser Bucht bedeutend verlangsamt sein, was zu einer Konzentration der Liquorbestandteile führt. Dieselbe könnte für die intensivere Osteoid- bzw. Knochenablagerung in dieser Gegend des Tractus verantwortlich sein (Fig. 1).

Um die dritte Frage beantworten zu können, u. zw. was primär ist, die Knochenablagerung oder die Nervenatrophie haben wir solche Fälle ausgesucht, wo es bestätigt war dass es sich um eine primäre Atrophie des Nerven handelte. Einen solchen Fall haben wir an den histologischen Schnitten prüfen können wo die Nervenöffnungen offen geblieben sind, obwohl der ganze Akustikus 40 Jahre vor dem Tode verengung. Als den zweiten Fall können wir denjenigen von Fisch (1971) erwähnen, wo etwa 3 J. nach der Atrophie des N. VIII der Meatus noch immer breit blieb obwohl er nur den N. facialis enthielt. Auch diejenigen Fälle, wo die Osteoidapposition um die Nervenbündel in die Richtung des inneren Gehörganges hinein stattfindet, sprechen für eine primäre Knochenapposition. Man könnte auch diejenigen Fälle hinzufügen, wo sich knöcherne Zylinder um die Arterienäste im Fundus bilden und die wir sowohl an makro- als auch an histologischen Präparaten gefunden haben. Die Arterien waren funktionsfähig und von knöchernen Manschetten, die in die Fundusrichtung hervorragten, umgeben.

Wir haben in unserem Institut, im Rahmen einer Magisterarbeit (Ciglar I) folgendes Experiment durchführen lassen. An 31 Ratten (17 junge von 2-3 Monaten und 14 alte von 1-2 Jahren) wurde der rechte bzw. der linke N. opticus durchschnitten. Drei Monate nach der Operation wurden die Tiere geopfert und

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III Ist die Knochenablagerung die Ursache der Folge der Atrophie und des Schwundes der Nervenfasern?

IV Wie verhält sich ein knöcherner Nervenkanal wenn der Nerv im Experiment durchtrennt wird?

Um die erste Frage zu beantworten untersuchten wir 1) andere Bereiche der Schädelbasis, wo die Nervenfasern ebenfalls durch ein System von siebförmigen Öffnungen hindurchtreten u. zw. die beiden vestibulären Arcen und die Lamina cribrosa, 2) die Nerven bzw. Gefäßkanäle der Schädelbasis u. zw. den Canalis rotundus, das Foramen ovale, das Foramen stylomastoideum, den Canalis hypoglossi und das Foramen spinale und 3) die Foramina intervertebralia in bezug auf die Osteophytenbildung der Wirbelkörper.

Aufgrund dieser Untersuchungen die alle

im Rahmen unseres Institutes durchgeführt wurden konnten wir folgendes feststellen.

1) Die Knochenablagerung findet auch in den vestibulären Arcen statt u. zw. intensiver in der sakulären als in der utrikulomampulären Gegend, was sowohl an mazerierten Präparaten als auch an histologischen Schnitten bestätigt werden konnte. Diese Knochenablagerung könnte für einige Alterserscheinungen im Gleichgewichtsorgan verantwortlich sein (Presbyostasis). Kelemen hat bei einem toten geborenen Hydrocephalus (ohne Anomalien des Ohres) Exostosen im inneren Gehörgang an die Äste des N VIII für den Sacculus gefunden. Er äußert die Möglichkeit, dass die selben Störungen in der vestibulären Funktion hervorrufen könnten. Die Öffnungen der Lamina cribrosa verengen und verschließen sich allmählich mit dem Alter in posterolateraler Richtung, was für die Altersstörungen des Riechsinnes verantwortlich sein könnte (Presbyosmie).

2) Pečina M. hat die Altersveränderungen des Canalis rotundus und des Foramen ovale am Kellbein, durch welche der II bzw. III Trigeminusast durchtreten, am grossen Material unseres Instituts untersucht. Er konnte feststellen, dass der Canalis rotundus bei den Frauen im Durchschnitt enger ist und dass dessen Durchmesser mit dem Alter in beiden Geschlechtern um etwa 0,4 mm abnimmt (von 3,37 im 11–20. Lebensjahr auf 2,97 im 80–90 Lebensjahr). Das Foramen ovale ist enger auf der rechten Seite und im weiblichen Geschlecht und verengt sich ebenfalls allmählich in beiden Geschlechtern mit dem Alter. Diese Altersverengung der beiden Öffnungen könnte für die Trigeminusneuralgie im mittleren und höheren Alter verantwortlich sein.

Večerina (1972) hat an demselben Material auch eine signifikante Verengung des Foramen stylomastoideum, des Foramen spinale und des Canalis N hypoglossi mit dem Alter gefunden. Das Foramen stylomastoideum ist bei den Frauen im Durchschnitt enger als bei den Männern und wird im Alter in beiden Geschlechtern progressiv enger. Der Prozen-

Mrs Krawpskić (Reply) to Mr Jangkees. With my remark concerning the narrowing of the facial canal and Bell's palsy I meant that may be this narrowing could cause the spasm of the arteries in the foramen which is considered by some as being the cause of the Bell's palsy.

To Mr Fisch. I am very grateful to Mr Fisch for

his discussion and for his remark about the contents of protein in the liquor in the region of the internal auditory meatus. His investigation and his idea of decompression of the internal auditory meatus are in complete accord with our idea concerning this problem.

die Nervendurchtrittsstelle an der operierten mit derjenigen an der nicht operierten Seite, die uns als Kontrolle diente verglichen. Obwohl der Nervenkanal auf der einen Seite leer war fand doch keine Knochenapposition statt. Bei alten Ratten waren die Nervenkanäle auf der operierten und nicht operierten Seite gleich gross. Bei jungen Ratten fand eben falls keine signifikante Verengung statt obwohl eine Verengungstendenz angedeutet wurde. Dies würde etwa den neuesten experimentellen Untersuchungen von Sarnat Shanedling über Verengung der Orbita nach der Exenteration bzw. der Enukleation des Augapfels entsprechen.

Weitere experimentelle Untersuchungen in diesem Sinne sind im Gange.

RÉSUMÉ

Supposant que la cause de la presbycusis est une apposition osseuse progressive dans la région du crible spirale suivie d'une atrophie des fibres nerveuses nous avons examiné aussi les autres trous nerveux de la base du crâne. Nous avons pu constater qu'à l'âge avancé on trouve une apposition osseuse aussi dans la région des autres trous de la base résultant en leur étrecissement. Ce procédé est responsable pour certaines manifestations dans la fonction de ces nerfs. Nous avons essayé aussi de prouver ces faits par des expériences sur des rats.

SUMMARY

Supposing that the cause of presbycusis is a progressive bone apposition in the region of the spiral tract with consecutive atrophy of the nerve fibres, we investigated also the other nerve openings in the base of the skull. We have found that in advanced age bone apposition also takes place in the region of other nerve openings which become narrower. This fact is also responsible for some manifestations in the function of these nerves. We also tried to prove our point in some experiments on rats.

LITERATUR

- Alexander G. 1902. Zur Frage des postembryonalen Wachstumes des menschlichen Ohrlabyrinthes. *Anat. Hefte* 63: 569.
Fisch, U. 1971. Degenerative changes of the arterial vessels of the internal auditory meatus during the process of aging. *Acta Otolaryng. (Stockh.)* 73: 1-6.

- Klemen, B. & Linthicum, F. H. 1969. Labyrinthine otosclerosis. *Acta Otolaryng. (Stockh.)*, Suppl. 23.
Krmpotić Nemančić, J. 1960. Bony cuffs around the branches of the internal auditory artery at the bottom of the internal auditory meatus. *Bull. Sci. RSFJ (Zagreb)* 4: 78.
— 1963. Presbycusis and Presbyosmia as Folgen eines analogen biologischen Prozesses. *Bull. Sci. RSFJ (Zagreb)* 8: 135.
— 1964. Presbyostasis. Abnahme der Reaktionsgröße des Gleichgewichtsapparates mit fortschreitendem Lebensalter sowie die Ursache dieser Erscheinung. *Bull. Sci. RSFJ (Zagreb)* 9: 19.
— 1968. Presbycusis and retrocochlear structures. *Int. Audiol. London Congr.* 7: 4.
— 1971. A new concept of the pathogenesis of presbycusis. *Arch. Otolaryng. (Chic.)* 91: 161.
— 1972. Macroscopical and mikroskopical changes in the bottom of the internal auditory meatus. *Acta Otolaryng. (Stockh.)* 73: 34.
Meyer M. 1923. Histologische Studie über den Gefäßreichtum insbesondere über die Entstehung der sog. durchbohrenden Kanäle und Stäbchen Gebilde in der knöchernen Labyrinthkapitel von menschlichen Föten und Jungkindern. *Z. Anat. Entwicklungsgesch.* 69: 521.
Bercer, A. & Krmpotić Nemančić. 1951. Über die Ursache der progressiven Altersschwerhörigkeit (Presbycusis). *Acta Otolaryng. (Stockh.)*, Suppl. 143.
Večerin, S. 1972. Morfološke promjene na nervu *otoclorina* baze lubanje tokom starenja. Thesis (Magister of biological sciences).

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DISCUSSION

L. B. H. Jongheer: Je félicite Mme Krmpotić de la ténacité avec laquelle elle continue de fortifier sa théorie et j'accepte aussi comme très probable que le transylvanostoidien d'vient plus petit avec l'âge. Hélas, je ne peux pas la suivre. Si elle pense que ce phénomène pourrait être à base de la paralysie de Bell, dans cette maladie le nerf n'est pas comprimé par l'os mais par la gaine nerveuse qui, dans la partie verticale ne contient pas de fibres élastiques.

U. Fisch: I would like to support one observation of Mrs Krmpotić concerning the possible role of a disturbance in the circulation of the cerebro-vascular field in the inner meatus in the production of deafness. We have measured the protein content of the meatal c.s.f. in progressive sensor-neural deafness and found an abnormal value sometimes of more than 80 mg. This observation has a practical significance since we are able to-day to improve surgically the circulation of c.s.f. in the internal auditory meatus.

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SNORING AND PRESBYACUSIS

M. Pražić

From the E.N.T. Department University of Zagreb Zagreb Yugoslavia

Abstract. The pathophysiological mechanism of snoring is briefly explained. The results of spectroscopic analysis of snoring are given. The analysis has shown some standard types of snoring. The parallelism between snoring and noise is pointed out. The audiograms of all examined snorers showed the presbycusis reduction of hearing.

Snoring is such a common and harmless phenomenon that one wonders whether to categorize it as a pathological manifestation or not.

A small number of scientific papers written on the phenomenon of snoring have now been published. The only one I came across was the Presidential Address under the title "Snoring" given by J. G. Robin at the Laryngological Section of R.S.M. in Great Britain in 1967 but he only dealt with secondary pathological complications and troubles caused by snoring. A number of other authors discussed this phenomenon in rather general terms, considering it as being a plain fact that should be prevented. Not a single one tried to find the real cause of snoring.

Nevertheless, a number of paramedical papers written on this phenomenon can be found.

The usual opinion is that mainly human beings snore especially those above 40 years. But the fact is that younger males and even elderly women snore too. Some people snore stronger some milder. Some snore during the whole night, some periodically. Some snore loud, and some very loud.

Special societies of snorers have been founded in several countries concerned solely

with the problem how to construct such a device which would awake the snorer immediately when he starts snoring. But until the present day no such device has been constructed that would prevent snoring.

At the meetings of the Collegium in Edinburgh and Lyon some years ago I talked about the pathophysiological mechanism of snoring. This time I will talk about the results of the audiological analysis of snoring and its acoustic qualities.

Intensity of snoring depends on the degree of dryness both of the mucous membrane of the soft palate and of the uvula. Dryness of the mucous membrane of the soft palate and of the uvula depends on the lessening of secretion of mucin from the seromucinous glands in the submucous layer of the upper respiratory tract. The lessening of secretion of mucin is caused by the changed or interrupted neurovegetative impulses of the glands. Since the secretion of mucin falls gradually the seromucinous carpet at the epithelial surface becomes serous, i.e. watery. During the respiration the strong and warm stream of air dries and stiffens the soft palate and uvula. They lose their elasticity and start vibrating. First they vibrate moderately and with mild intensity. Later on, the intensity of vibrations grows stronger.

The measuring of the intensity of snoring shows a relatively broad variety. There are snorers whose intensity of snoring reaches the level of 70 dB. (Measuring was performed at the distance of 25 cm from the mouth of the

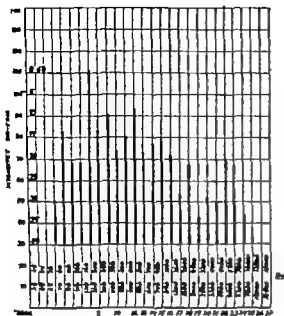


Fig. 1

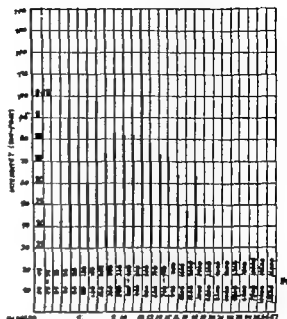


Fig. 2

) Some snorers even reach the level of dB.

The frequency analysis of snoring shows interesting results. The mechanism of snoring is very similar to the mechanical elements of the esophageal voice. Therefore the frequency and the amplitude elements of snoring have similar components.

There are several types of snoring. The first is sonorous snoring that ranges from 60–10 000 c.p.s. (Fig. 1). The second type is not so sonorous as the first and therefore this seems deep. Here the range of frequencies reaches from 60 up to 2 000 c.p.s. only (Fig. 2).

The third, fourth and fifth type are more sonorous. Here the range of frequencies covers the field from 200–8 000 c.p.s. They are very similar but in many details they are different (Figs. 3–5).

The effects of sonority, depth and height of snoring depend not only on the range of frequencies but also on the amplitude variations of single segments of frequencies. These variations reach as much as 30 dB in some

(there were altogether 17 snorers), were male persons above 60 years. Only the range from 60–10 000 c.p.s. was analysed. The analysis did not cover the infrasound range, or the range of vibrations. The reason for this lack of analysis lies in the technical field. It is very dif-

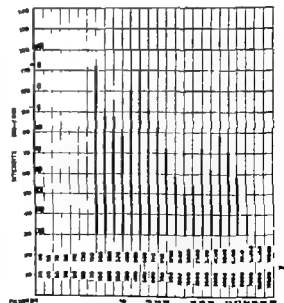


Fig. 3

All the cases of snoring that I examined

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The measuring of the intensity of snoring shows a relatively broad variety. There are snorers whose intensity of snoring reaches the level of 70 dB (Measuring was performed at the distance of 25 cm from the mouth of the

nal auditory ductus. In this way one can explain some of the presbyacoustic reduction of hearing.

If one accepts that the majority of elderly snore and if one takes into consideration that the snorer snores every night for several hours, the intensity of snoring being 80 or above 80 dB, combined with the vibrations, there is a completely new etiological factor for presbycusis. The snorer snores every night for years, which means that he is exposed to a kind of noise. Therefore a snorer can be compared to a worker in a noisy factory.

Amongst the etiologic factors that can cause presbycusis one must therefore take into consideration snoring as a primary factor.

RÉSUMÉ

On donne un exposé bref du mécanisme du ronflement. Puis on donne des résultats de l'analyse spectrographique du ronflement concernant les variations en fréquence et en amplitude. L'analyse montre quelques types standardisés du ronflement. On indique le parallélisme entre le ronflement et le bruit et on souligne les faits concernant l'effet nuisible du bruit sur l'ouïe. On présente des audiogrammes tonaux des ronfleurs et, se basant sur ces audiogrammes, on montre une liaison étiologique entre le ronflement et la presbycusis.

ZUSAMMENFASSUNG

Der pathophysiologische Mechanismus des Schnarchens im kurzen ist dargestellt. Nachdem sind die Resultate der spektroskopischen Analyse des Schnarchens mit den Frequenz und Amplitude-Variationen gezeigt. Analyse zeigt einige Standard-Typen des Schnarchens. Der Parallelismus zwischen Schnarchen und Lärm wird betont sowie einige Tatsachen über den schädlichen Effekt von Lärm auf das Gehör. Eine Serie von Audiogrammen vom Schnarchen wird gezeigt und auf Grund dieser Audiogramme wird eine etiologische Kausalität des Schnarchens mit der Entstehung der Presbyakusis angenommen.

REFERENCES

- Krmpotić-Nemendić, J. 1972. Macroscopical and microscopical changes in the bottom of the internal auditory meatus. *Acta Otolaryng* (Stockh.) 73: 54.
- Prall, M. 1967. The quantitative measurement of mucus in the upper respiratory tract. *Acta Otolaryng* (Stockh.) 63: 264.
- 1963. The quantitative values of mucus in respiratory tract in normal and pathological conditions. *Acta Otolaryng* (Stockh.) 57: 377.
- Saxén, A. 1965. Pathologie und Klinik der Altersschwerhörigkeit. *Acta Otolaryng* (Stockh.), Suppl. 23.
- Schulzecht, H. F. 1955. Presbycusis. *Laryngoscope* 65: 402.
- 1957. Further observations on the pathology of presbycusis. *Arch Otolaryng* (Chic.) 60: 369.
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RELATION OF STRUCTURAL DAMAGE TO EXPOSURE TIME
AND INTENSITY IN ACOUSTIC TRAUMA

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Abstract In order to overcome the difficulty to obtain a survey evaluation of the entire cochlea as well as an ultrastructural examination of selected areas of the same ear we used a new technique for combined surface preparations and EMI studies without the risk of damage by mechanical preparation. Exposure intensities were varied from 100 to 140 dB and exposure times from 30 sec to one week. The relations between exposure time and extent or type of damage on one hand and between exposure intensity and damage on the other hand were worked out at different intensity levels. Above 120-130 dB exposure intensity direct immediate structural damage occurs regularly in various degrees, depending mainly on exposure intensity whereas in lower intensities delayed phenomena are predominant, depending mainly on exposure time.

In order to study appropriately the structural damage in acoustic trauma a new technique is used for the preparation of cochleas, allowing a total surface evaluation of all turns as well as light and electron microscopic examinations at any magnification of any desired area of the same specimen. The findings can easily be documented at every step and the risks of mechanical artefacts due to the preparation are reduced to a minimum.

The intact cochlea, opened only at the round and oval window, is fixed and embedded as a whole in a new low viscosity epoxy resin (Spurr). After polymerisation the embedded cochlea is cut in two halves with a 0.1 mm thick circular saw under a preparation microscope. From the two cochlear halves horizontal slices of each cochlear turn are excised and used directly for surface evaluation (Fig. 1). Selected areas of these cochlear turn

slices are then dissected out and used for semi-thick and ultrathin sections for conventional light and electron microscopic examination (Spoendlin 1972). A somewhat different procedure aiming at the same final goal has been reported by Bohne (1972) and by Ernst (1972).

In the present experiment 110 guinea pigs were exposed to a wide band noise at intensities between 110 and 140 dB and with exposure times between 30 sec. and 1 week. The animals were sacrificed either immediately after the exposure or after a survival time of 3 weeks and 3 months.

At exposure intensities of 130 dB severe structural alterations are found immediately after the exposure. Even after 30 sec exposures to 140 dB the organ of Corti was usually entirely missing in a small area of the lower second turn leaving only the bare basilar membrane with the spiral vessel and the tectorial membrane (Fig. 1). This zone of missing organ of Corti is sharply limited on both sides against regions of more or less intact structures showing different degrees of distortion. Quite frequently the organ of Corti is not actually missing but roughly disintegrated with ruptures of the reticular membrane or pillar heads, dislocation or expulsion of sensory cells, disconnection of the organ of Corti from the basilar membrane and the tectorial membrane and a complete loss of the tympanic lamina (Fig. 2). Such drastic primary alterations lead necessarily to a complete and permanent loss

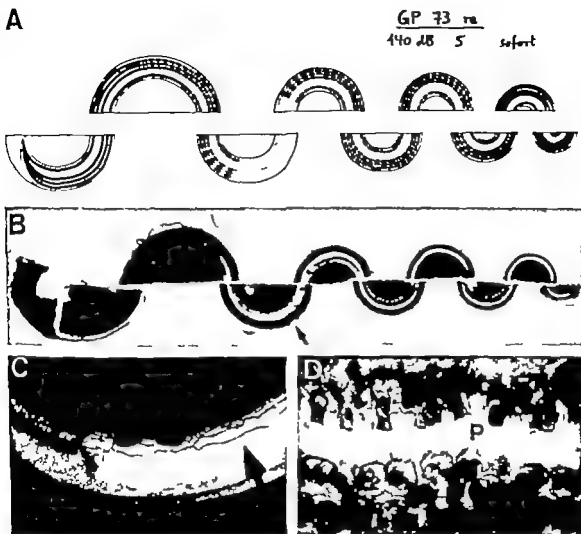


Fig. 1 (A) Diagrammatic representation of the surface preparation of the cochlea depicted in (B). Thick interrupted lines represent areas of distortion whereas thin interrupted lines represent areas of completely missing organ of Corti. (B) A total view of a surface preparation of a guinea pig cochlea immediately after an exposure to 140 dB for 5 minutes. That the organ of Corti is completely missing in the upper basal and lower second turn is clearly seen (arrow). (C) Higher magnification of a portion of the upper basal turn

of the cochlea in (B). In the region where the organ of Corti is entirely missing the spiral vessel is still clearly visible (arrow). There is an abrupt transition from entirely missing organ of Corti to a more or less intact organ of Corti on the left. (D) Detail of an organ of Corti showing heavy distortion, respectively disintegration. The outer hair cells (*H*) are irregularly swollen and distorted. The pillar heads (*P*) are for the greater part entirely missing. (Compare Fig. 2).

of all cellular elements of the organ of Corti. These alterations are obviously the consequence of a direct mechanical damage not only because of their appearance but also because of the very short exposure times of a few seconds needed to produce them.

A quite different type of lesion is found

after longer exposures to lower intensities, below 120 dB. Single outer hair cells degenerate amidst entirely normal cells, scattered over large distances. The primary changes are always confined to the sensory cells, which are swollen with a vacuolization of the cytoplasm and mitochondrial degeneration and are fl-



Fig. — Low magnification, electron microscopic picture of an organ of Corti in disintegration corresponding to the area depicted in Fig. 1 D. The pillar heads (P) are ruptured and the whole organ of Corti is burst open towards the endolymphatic space. The outer hair cells (OH) and the inner hair cells (IH) are heavily distorted and partly exposed in the endolymphatic space. The fine cytological elements as well

as the internal spiral fibres do not show much significant alteration. The tympanic lamina is almost entirely missing and only the spiral vessel (T) remains in place. The whole organ of Corti seems to be disconnected from the basilar membrane and the tabanula region. The tectorial membrane (M) is essentially normal.

nally completely disintegrated whereas the nerve endings still remain intact at this stage. Scattered degeneration occurs also to a much smaller degree spontaneously in normal animals, which means that a normally occurring process is merely enhanced by acoustic over

stimulation. For these changes as well as for the frequently observed swelling of the dendrites to the inner hair cells (Spöndlin, 1970, 1971) a purely mechanical mechanism is excluded and metabolic disturbance such as metabolic exhaustion is the most likely cause

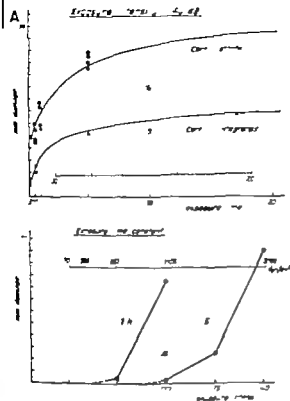


Fig. 3. (A) Diagram showing the relation between the extent of damage (in mm) and the exposure time at a constant intensity of 140 dB. The upper curve relates to the entire area of distorted organ of Corti whereas the lower curve relates to the area where the organ of Corti is entirely disintegrated or missing. After a very short initial rising phase the curves flatten out to become almost horizontal. If time scale is taken logarithmic (smaller inset scale) the curves become more linear (smaller inset scale) the curves become more linear, slowly rising lines (interrupted lines). (B) Diagram representing the relation between the extent of damage (in mm) to the exposure intensity at constant times of one hour (left curve) and 3 minutes (right curve). Above a certain critical level the curves rise with increasing steepness. If the exposure intensity is expressed in a linear manner as dyn/cm^2 as shown in the upper inset scale with corresponding sound pressure values, the curve (interrupted line) becomes almost straight.

At exposure intensities of 120–130 dB the structural alterations consist mainly of different degrees of distortion of the hair cells, which can be attributed to a mechanical as well as to a metabolic disturbance. The very typical outbending and fusion of the sensory hairs of the inner hair cells is found even after relatively mild exposures and usually extends

apicalwards far over the main damage zones as the only significant structural alteration (Spoendlin, 1971). Fusions of sensory hairs appear to be a rather general and unspecific response to many different types of damaging factors such as intoxication and genetic disturbance (Duvall & Wersäll, 1964; Ernston, 1971).

Following the noise exposure, the degeneration of damaged elements proceeds and reaches its final permanent status only after a longer period of time. In areas of disintegration only the bare basilar membrane remains, covered by a single layer of flat epithelial cells, the spiral vessel and the tectorial membrane. Areas of severe distortion end up as a collapsed group of supporting cells with entirely missing hair cells or with only the inner hair cells left. In places where only the inner hair cells remain there is no appreciable retrograde degeneration of the cochlear neurons, as was also observed by Ward & Duvall (1972) and which is in agreement with our earlier finding that 95% of the cochlear neurons are associated with the inner hair cells (Spoendlin, 1972). Scattered degeneration of outer hair cells leaves a normal organ of Corti with just a few missing outer hair cells. The only minor change to remain within an otherwise normal organ are the distorted sensory hairs of the inner hair cells. Swelling of dendrites to the inner hair cells and slight distortion of the outer hair cells are probably reversible to a certain extent (Spoendlin, 1971). Although the structural alterations become more pronounced as the process of degeneration proceeds with postexposure time the extent of damage observed immediately after high intensity exposures does not increase considerably. However, milder damage after moderate intensity exposures frequently shows up only after a certain time delay in the form of scattered hair cell degeneration.

For easier survey the main types of structural alteration are set out in a diagram which is the exact copy of the original surface preparation of the cochlea (Fig. 1) showing the

missing and disintegrated organ of Corti different degrees of distortion, and missing hair cells, and which is similar to the cochleogram used in classical surface techniques (Engström et al. 1966).

On the basis of our combined technique of evaluation of ultrastructural damage with reconstruction of the entire cochlea it is possible to compare the extent of structural damage with the intensity of time of exposure. The area of completely missing or disintegrated Corti and the area of distortion were used as structural parameters which can be accurately evaluated and certainly reflect the degree of cochlear damage.

There is a characteristic relation between the extent of immediate structural damage and different exposure times at constant exposure intensity (Fig. 3 A). At high intensities of 140 dB the curve flattens out to an almost horizontal course after a very short initial steeply raising phase. At lower intensities of 130 dB and less the raising portion of the curve extends over an increasingly longer time before it flattens out. If the time scale is logarithmic the curve becomes a straight line with a small raising angle similar to the TTS which grows linear in the logarithm of time (Ward, 1970).

The relation of damage to exposure intensity in dB at constant exposure times, on the other hand, shows a different characteristic of the curve, which above a certain critical intensity level rises with increasing steepness. If however exposure intensity is expressed in a linear way in terms of dyn/cm^2 the relation appears to become once more a nearly linear function (Fig. 3 B). In order to obtain an equal degree of cochlear damage, the exposure time must be increased exponentially when exposure intensity is decreased.

In conclusion, it is clearly demonstrated that direct mechanical destructions as well as metabolic exhaustion are competing factors in acoustic traumatic damage of the cochlea. Direct mechanical damage is usually irreversible and appears immediately after relatively

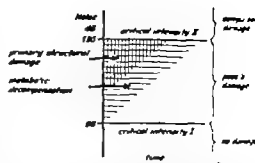


Fig. 4. Diagram representing the three intensity zones of sound in respect to its acoustic traumatic effect. Up to 90 dB (critical intensity I) no damage is to be expected. Between 90 dB and 140 dB metabolic decompensation occurs increasingly and even primary structural damage might appear to a certain extent. In this intensity zone the organ of Corti may or may not be damaged, at which the exposure time might be an important factor. Above the critical intensity II of 130 dB there is always heavy primary structural irreversible damage of a direct mechanical type even after very short exposures.

short exposures at high intensities above a certain level whereas metabolically induced damage is partly reversible, occurs after long exposures with moderate intensities and develops more slowly over a longer period of post-exposure time. Mechanical damage obviously depends on the mechanical resistance of the structure of the organ of Corti and therefore does not show much individual variation. The metabolically induced damage, however, exhibits great differences among the individual animals. Thus in respect of acoustic traumatic effects three zones of intensity can be delineated (Fig. 4). Up to about 90 dB the classical critical intensity level, practically no damage is produced. From 90 to 130 dB permanent acoustic traumatic damage mainly of the metabolic type may occur to a varying extent. Above 130 dB severe irreversible structural damage is unavoidable.

Exposure time and intensity do not seem to be equally responsible for structural damage. At higher intensity levels, exposure intensity is by far more decisive on the extent of damage than is exposure time, except for a short initial period where the time factor appears to be equally important. This initial pe-

nod is very short at high intensity exposures and becomes increasingly longer at lower intensities. Beyond this initial period the amount of damage seems to be mainly defined by the exposure intensity. Thus equal total energy does not seem to produce always the same amount of damage, which is in agreement with recent findings of Ward & Duvall (1972).

RÉSUMÉ

Pour surmonter la difficulté d'obtenir une évaluation globale de toute la cochlée ainsi bien qu'un examen au niveau d'ultrastructure nous avons adopté une nouvelle méthode permettant une combinaison entre préparation de surface et de microscopie électronique. Dans les expériences des intensités de 100 à 140 dB et des temps d'exposition de 30 secondes à une semaine ont été employées. Aux intensités au delà de 125-130 dB un dommage morphologique direct et immédiat est régulièrement observé, dépendant sur tout de l'intensité de l'exposition acoustique, tandis qu'aux intensités plus faibles des phénomènes retardés sont prédominants et dépendent surtout de la durée de l'exposition acoustique.

ZUSAMMENFASSUNG

Mikroelektronische Übersicht über die ganze Ohr- und gleichzeitiger ultrastruktureller Untersuchung ausgedehnter Abschnitte wurde ein neues Verfahren verwendet, das eine Kombination von Oberflächenpräparation und Elektronenmikroskopie lautet. Zur Schallexposition wurden Intensitäten von 100-140 dB mit verschiedener Dauer von 30 Sekunden bis einer Woche verwendet. Die Beziehungen zwischen Ausdehnung oder Art des Schadens zur Dauer und der Intensität der Schallexposition wurden untersucht. Bei Intensitäten oberhalb 125-130 dB sind direkte strukturelle, meist irreversible Immediatschäden schon bei kurzen Expositionzeiten die Regel, während bei schwächeren Intensitäten Spätveränderungen vor herrschen, deren Auftreten und Ausmass entscheidend von der Expositionsdauer abhängen.

REFERENCES

- Bohne, R. A. 1972. Location of small cochlear lesions by phase contrast microscopy prior to thin sectioning. *Laryngoscope* 82, 1.
- Dovall, A. J. & Wernall, J. 1964. Site of action of streptomycin upon inner ear sensory cells. *Acta Otolaryng* (Stockh.) 57 581.
- Engström, H., Ades, H. W. & Anderson, A. 1966. *Structural Pattern of the organ of Corti. A systematic mapping of sensory cells and neural elements*. Almqvist & Wiksell/Gebens Förlag AB Stockholm.
- Ernstson, S. 1972. Cochlear morphology in a strain of the waltzing guinea pig. *Acta Otolaryng* (Stockh.) 71 469.
- 1972. Cochlear physiology and hair cell population in a strain of the waltzing guinea pig. *Acta Otolaryng* (Stockh.), Suppl. 297.
- Lin, H. J. & Melnick, W. 1971. Acoustic damage of the cochlea. *Arch Otolaryng* (Chic.) 94 294.
- Spoendlin, H. 1970. Ultrastructure of the peripheral nervous system and sense organs. In *Atlas of normal and pathologic anatomy* vol. VIII. Georg Thieme Verlag, Stuttgart.
- 1971. Primary structural changes in the organ of Corti after acoustic overstimulation. *Acta Otolaryng* (Stockh.) 71 166.
- 1972. Innervation densities of the cochlea. *Acta Otolaryng* (Stockh.) 73 235.
- Ward, D. W. 1971. Biochemical implications in auditory fatigue and noise induced hearing loss. In *Biochemical mechanisms in hearing and deafness* (ed. M. M. Paparella). C. C. Thomas, Springfield Ill.
- Ward, D. W. & Duvall, A. J. 1972. Behavioral and ultrastructural correlates of acoustic trauma. *Ann Otol* 80 881.

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DISCUSSION

H Engström. Mr Spoendlin's findings are in agreement with what many other scientists are observing with different kinds of technique. One interesting problem is the occurrence of hearing loss and little modification of the organ of Corti as recently discussed in Chicago. Many scientists are studying the effect of noise and only by a systematic attack with many different approaches can we solve what happens after noise exposure. In Uppsala we work very much along similar lines as Mr Spoendlin and we also use scanning electron microscopy extensively (6 figures shown).

E. Bocca. I should like to know how Mr Spoendlin explains the apparently conflicting evidence resulting from animal observations and from audiometric observation of human subjects exposed to noise.

For extremely high intensities, maximum damage may be a matter of seconds or minutes and cannot possibly be increased by a longer period of exposure. However for critical intensities, total exposure time, or even more so, fractioning of the total exposure time seems to be more important than intensity causing, or preventing noise-induced loss of hearing. Whenever we allow the acoustic cells time enough to rebuild their metabolic reserves, we may even expose

them to short duration high intensity noises with no apparent permanent damage.

K. H. Vosteen. Die systematische Technik von Dr. Spoendlin erlaubt mechanische Schäden von Stoffwechselerschöpfung in den Sinneszellen nach akustischem Trauma besser zu unterscheiden. In dieser Beziehung habe ich 3 Fragen: 1) Haben Sie auch längere Zeit (mehr als 2 Wochen) mit geringerer Intensität (90-100 dB) beschallt? 2) Haben Sie auch mit unterbrochenen Tönen oder Geräuschen beschallt? 3) Wie sind die Resultate bei unterschiedlicher Überlebenszeit?

D. Hilding. Since we learned of the possibility of using plastic-embedded tissue for surface preparations from Mr. Wersäll, we have found the technique to be very useful. My question concerns storage of your beautiful "Loch Ness" displays. Do you have a way to permanently store them? I would suggest replacing glass coverlips and slides with epoxy ones—and mounting in epoxy. This will permit recovery of interesting areas for electron microscopy later.

E. Borghesen. Les très belles slides du Mr. Spoendlin ont montré la nécrose des groupes bien différenciés de cellules sensorielles; je demande s'il y a une prédisposition anatomique et, éventuellement, quelle est.

G. Lidén. Would you care to comment further on your experience of the equal energy principle. According to your results would you say that this principle is not generally valid or that it is useful only in certain intensity ranges. Finally have you done any experimental studies on the effect of impact or impulse noise?

A. C. Hilding. 1) Did Mr. Spoendlin use pure tone or white noise in his experiments? 2) If pure tone did the highly localized lesions produced by lower intensity sound correspond in localization with the pitch of the tone? 3) Mr. Spoendlin referred to these localized lesions as a result of metabolic change. Does he have an explanation for the sharpness of the localization?

H. Spoendlin (Reply) to Mr. Engström. The reason why we preferred this technique to scanning electron microscopy is because we consider it very important to be able to survey the entire cochlea in a surface manner and to study every selected part of the same cochlea in light and electron microscopy. In scanning electron microscopy you are forced to break open the cochlea, which implies considerable tissue losses and mechanical artefacts. Further evaluation of the same material in light and transmission electron micro-

scopy is limited because of preparation artefacts. It is also our experience that we always find a marked TTS after lower intensity exposures without any significant structural alterations in the cochlea. The first alteration to be observed after low intensity exposure is scattered degeneration of outer hair cells. Sensory hair alterations occur mainly at the inner hair cells but only at somewhat higher exposure intensities.

To Mr. Bocca. There is not only a difference in noise-induced damage susceptibility between animals and humans but also between different animals. The chinchilla seems, for instance, to be much more susceptible than the guinea pig (Ward & Durrill, 1972). The relatively greater importance of exposure intensity for noise-induced cochlear damage was demonstrated for high intensity exposures, where extreme primary damage occurs within a short time after which further increase of exposure time produces only a very small increase of structural damage. At lower exposure intensities the initial rising phase of the damage-exposure-time curve extends over a longer time before it levels out. At this initial rising phase the exposure time seems to have considerable importance and the principle of equal energy might just be valid for this initial phase. It is known that the hearing of "noise workers" who are exposed over years to the same noise does not deteriorate further after a certain number of years.

To Mr. Vosteen. Low intensity exposures of 110 dB were given for 1 week without any pronounced cochlear damage whereas exposures with interrupted tones in form of speech at peak intensities of 140 dB produced much greater damage than a steady noise exposure at 140 dB. So far no combination of noise exposure with hypoxia has been undertaken.

To Mr. Hilding. Our surface preparations are stored in immersion oil where they are always ready for examination and further evaluation.

To Mr. Borghesen. It is indeed a striking fact that even after very short high intensity exposures a sharply delimited lesion is produced, predominantly by direct mechanical effects of the sound, in the lower second or upper basal turn. The reason for this localization might be a greater mechanical susceptibility of the organ of Corti in this region or, even more likely, a maximum of volume displacement in this region.

To Mr. Lidén. This question has been answered in response to Bocca and Vosteen.

ALKALINE AND ACID PHOSPHATASE ACTIVITY IN POSTAURICULAR SKIN AND CHOLESTEATOMA EPITHELIUM

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Abstract. Results are reported on quantitative and electrophoretic analyses of alkaline and acid phosphatase activity in cholesteatoma epithelium and postauricular skin. The activity of these two enzymes was significantly higher in the former than in the latter. Only one isoenzyme band with a mobility of α -globulins was found for alkaline phosphatase and another with a mobility of α -globulins for acid phosphatase in both tissues. The results prove that acid phosphatase plays an important part in keratinization and that the earlier reports of absence of alkaline phosphatase in squamous epithelium were due to the inhibitory effect of cysteine.

Studies on the histochemistry of cholesteatoma epithelium have increased recently since mechanical factors or epithelial immigration alone are insufficient to explain the development of cholesteatomatous disease. Keratinization as such has also become a widely studied subject because the abundant formation of keratin in cholesteatoma is one of its characteristic features (Jarrett et al. 1959).

Release of the hydrolytic enzymes in the granular cell layer determines mainly the type of keratin produced (Jarrett & Spearman 1967). The activity of acid phosphatase is specially noticeable in the granular cell layer and it is the only enzyme that can be demonstrated in great quantities also in the horny layer (Palva et al. 1970). On the other hand, no alkaline phosphatase activity has been found in the keratinizing squamous epithelium though it can be readily demonstrated in the capillary endothelial cells (Wells, 1960; Harris, 1962; Palva et al. 1970).

We have recently shown (Palva et al., 1972) that there is, nevertheless, some alkaline phosphatase activity both in cholesteatoma epithelium and in postauricular and ear canal skin. This activity could be demonstrated by removing the inhibitory effect of cysteine by blocking the active sulphhydryl groups with iodoacetic acid. Histochemically however no distinct difference was shown to exist between cholesteatoma epithelium and skin.

We now report on our extended studies using electrophoretic separation of these two enzymes from the cholesteatomatous membrane and skin homogenates and on quantitative determinations of acid and alkaline phosphatase total activity in these two tissues.

MATERIAL AND METHODS

Strips of postauricular skin were removed from 20 patients with cholesteatomatous ear disease undergoing surgery. Cholesteatoma epithelium was also removed and both tissue specimens were transported on gauze strips over blocks of ice to the laboratory. The tissue specimens, as well as the patient's serum, were stored at -25°C before analyses. For enzyme analysis, the tissue specimens were thawed and homogenization was made in physiological saline with a glass homogenizer at $+4^{\circ}\text{C}$. An amount of 1-2 ml of saline was used per 1 g of fresh tissue. The homogenate was centrifuged at 40 000 g for 20 min and the clear supernatant was used for quantita-

Table I Phosphatase activity in serum ($\mu\text{mol/min} \times \text{l l}$)

20 cases	Phosphatase		Inhibition %	
	Alkaline	Acid	Tartrate	Formaldehyde
Mean	43.4	4.60	16	84
S.D.	37.7	3.29	16	20

tive phosphatase determinations and for electrophoresis. Some quantitative measurements were made from a few specimens from the homogenate itself.

Electrophoretic separation of isoenzymes was carried out using a Wicke apparatus at $+12^\circ\text{C}$. For alkaline phosphatase analysis 1% agar (Difco special agar) in 0.1 mol/l Veronal buffer pH 9.5 was prepared. For acid phosphatase 1% agar was made in 0.05 mol/l citrate buffer pH 6.2. Electrophoresis was performed for 20 min with a current of 30 mA per slide.

The alkaline phosphatase fractions were stained using the methods of Rawstron (1971). β -naphthyl phosphate was used as substrate in 0.1 mol/l carbonate buffer at pH 10 while Fast Blue RR was used as diazo salt. The incubation period was 30 min in room temperature. The fractions of acid phosphatase were stained using the method of Smith & Hitchy (1968): 10 mg of α -naphthyl phosphate and 10 mg of Fast Blue RR in 10 ml 0.1 mol/l acetate buffer at pH 5.0. The incubation period was 2 h in room temperature.

Quantitative determinations of phosphatase activity were made after the method of King (1965). For acid phosphatase the substrate phenyl phosphate was dissolved in citrate buffer pH 4.9 and for alkaline phosphatase in carbonate buffer at pH 10.0. King's methods were also followed in the inhibition tests for acid phosphatase using tartrate and formaldehyde.

RESULTS

The results of alkaline and acid phosphatase activity tests in serum are shown in Table I

together with the inhibition tests for acid phosphatase. Some individual values on the high side appeared but the means are well within the normal range. The inhibition tests show that most of the acid phosphatase is formaldehyde labile (84%) while tartrate has a slight effect only (16%).

Table II shows the corresponding quantitative analyses of the supernatant of the fresh tissue homogenates prepared from cholesteatoma epithelium and of postauricular skin. In a few specimens no alkaline phosphatase activity could be shown but in the majority a small amount of activity was demonstrated. The mean figure for cholesteatoma epithelium was $0.22 \mu\text{mol/min} \times \text{g}$ and for postauricular skin $0.07 \mu\text{mol/min} \times \text{g}$. Student's *t*-test showed this difference to be significant at the $p=0.02$ level.

Acid phosphatase activity was constantly demonstrated in all analysed homogenates and the mean figures for cholesteatoma and skin were 1.98 and $0.67 \mu\text{mol/min} \times \text{g}$, respectively. Two values in the cholesteatoma group were markedly elevated (cases 16 and 19). The *t*-test showed a marginally significant difference between the averages ($p=0.05$).

Some analyses were also made of the homogenates without centrifugation and, in general, the measured activity increased in varying amounts. This was apparently due to the structure-bound lysosomal enzymes which do not remain in the supernatant after centrifugation.

Table II Phosphatase activity in fresh tissue homogenates ($\mu\text{mol/min} \times \text{g}$)

20 cases	Phosphatase		Inhibition %	
	Alkaline	Acid	Tartrate	Formaldehyde
<i>Cholesteatoma epithelium (20 cases)</i>				
Mean	0.24	1.98	82	37
S.D.	0.27	2.56	22	20
<i>Postauricular skin (20 cases)</i>				
Mean	0.07	0.63	90	35
S.D.	0.07	0.41	7	20



Fig. 1 Electrophoretic analysis for alkaline phosphatase of serum (S), cholesteatoma epithelium (C) and postauricular skin (PS) in case 9. Moderate activity

is seen in the area of α_2 -globulins for cholesteatoma while very weak bands appear in serum and skin. Control stains without substrate were negative.

The inhibition tests showed similar results in cholesteatoma epithelium and skin homogenates. Both were markedly resistant to the action of formaldehyde (63 and 65% respectively) while tartrate destroyed most of the enzyme (82 and 90% respectively).

In electrophoretic analyses the alkaline phosphatase activity was shown in the form of one rather weak band which migrated anodically from the application point with the mobility of α_2 -globulins. The acid phosphatase activity appeared as one single strong band cathodically from the application point, having the mobility of β_1 -globulins. The control tests without substrate were negative.

COMMENT

We have earlier demonstrated (Palva et al., 1970) in various enzyme studies that cholesteatoma epithelium has a lively metabolic activity and the present study shows that activity for alkaline and acid phosphatase is significantly higher in cholesteatoma as compared with postauricular skin. Our recent (Palva et al., 1972) histochemical demonstration of alkaline phosphatase activity in these tissues is here corroborated quantitatively and electrophoretically. This enzyme activity which was earlier thought to be absent from cholesteatoma epithelium and skin, is normally inhibited

by cysteine, which appears in the sulphhydryl groups of the keratinizing epithelium and is missed histochemically if special methods are not resorted to. In respiratory type of epithelium alkaline phosphatase appears both in the epithelial cells and in the ciliae and it can be speculated that inhibition of alkaline phosphatase activity in squamous epithelium is one of the features necessary for keeping the keratinization process going.

Acid phosphatase is one of the most active hydrolyzing enzymes which are released at the level of the granular cell layer. It assists in the breakdown of phospholipids whose energy can be utilized in the keratinization process. In cholesteatoma epithelium, the enzyme activity is marked and continues into the horny layer indicating a strong keratinization process. In normal skin, acid phosphatase activity does not extend into the horny layer and the breakdown of phospholipids may be incomplete. These histochemical observations are corroborated by the present quantitative assessment.

In this study we were particularly looking for such acid phosphatase isoenzyme bands as might be different in cholesteatoma epithelium and skin. However no such bands appeared and a quantitative difference only was observed.

There appeared to be a clear difference in

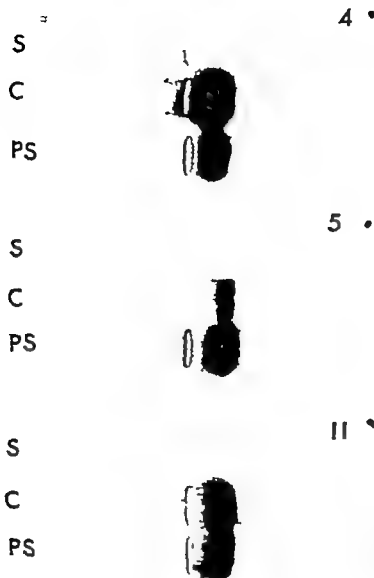


Fig. 2. Electrophoretic analysis for acid phosphatase of serum (S), cholesteatoma epithelium (C) and postauricular skin (PS) of cases 4, 5 and 11. Marked activity appears in the β -region with notable individual differences. Control stains without substrate were negative.

the origin of serum and tissue acid phosphatases. In serum they originate mainly from the red cells while in the cholesteatoma epithelium and skin the prostatic-type acid phosphatase dominates.

Cholesteatoma epithelium generally arises from the epidermis at the perforation margin in the form of ingrowing migrating epithelium. In some cases the squamous epithelium retains its normal characteristics of tympanic membrane outer surface in others it is aggressive, tumour-like cholesteatoma. The lat-

ter need by no means be associated with pressure keratinization since the epithelium retains its characteristics even on smooth open surfaces. It is probable that a definite enzymatic change in skin has developed which is responsible for the behaviour of cholesteatoma. There is earlier evidence of increased esterase (Palva et al., 1971) and collagenase (Abramson, 1969) activities in addition to the present evidence of increased phosphatase activity. However the exact process and the many other factors involved remain unknown for the present.

RÉSUMÉ

L'activité de la phosphatase acide et alcaline a été étudiée dans la membrane cholesteatomateuse et dans la peau avec des méthodes histochemiques. Une partie des biopsies a été homogénéisée et analysée avec des méthodes électrophorétiques. On discute les résultats.

ZUSAMMENFASSUNG

Die Aktivität der sauren und alkalischen Phosphatasen wurden quantitativ und elektrophoretisch an Homogenaten vom Cholesteatomsepithel und postaurikulärer Haut gemessen. Aktivität für beide Enzyme war bedeutsam größer im Cholesteatomsepithel. Nur ein Isoenzymband wurde demonstriert mit Mobilität von α_2 -Globulin für alkalische und β_2 -Globulin für saure Phosphatase. Saure Phosphatase hat einen wichtigen Teil in dem Keratinisationsprozess und die früheren Mitteilungen über den Mangel an alkalischen Phosphatasen sind durch den hemmenden Effekt von Cystin verursacht.

REFERENCES

- Abraham, M. 1969 Collagenolytic activity in middle ear cholesteatoma. *Ann Otol* 78 112.
 Harris, A. J. 1962. Cholesteatoma and chronic otitis media. The histopathology of ossous and soft tissues. *Laryngoscope* 72 954.
 Jarrett, A., Spearman, R. I. & Hardy, J. A. 1959. The histochemistry of keratinization. *Brit J Derm* 71 277.
 Jarrett, A. & Spearman, R. I. C. 1967. Keratinization. *Derm Digest* 6 43.
 Klig, J. 1965. *Practical clinical enzymology*. Van Nostrand, London.
 Paha, T., Palva, A. & Dammert, K. 1970. Middle ear mucosa and chronic ear disease. II. Enzyme studies. *Arch Otolaryng* (Chic.) 91 50.
 Paha, T., Rannio, V., Forsén, R. & Palva, A. 1971. Esterases of postauricular and ear canal skin, compared with cholesteatoma epithelium. *Acta Otolaryng* (Stockh.) 72 329.
 Paha, T., Palva, A. & Dammert, K. 1972. Alkaline phosphatase activity in cholesteatoma epithelium and skin. *ORL* 34 153.

- Rawstron, J. R. 1971. Rapid electrophoresis of alkaline phosphatase isoenzymes. *Clin Chim Acta* 52 303.
 Smith, J. K. & Whitby, L. H. 1965. The heterogeneity of prostatic acid phosphatase. *Biochim Biophys Acta* 151 607.
 Wells, G. C. 1960. Esterases in normal human skin and in chronic granulomata. In *Progress in the biological sciences in relation to dermatology* (ed. A. Rook) pp. 120-134. Cambridge University Press, London.

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DISCUSSION

H. H. Neumann. Das von Herrn Paha festgestellte Vorkommen alkalischer und saurer Phosphatasen in der Cholesteatommatrix erinnert mich sehr an Befunde, die kürzlich Dr. J. Herrmann, ein Mitarbeiter der Münchner Klinik bei der experimentellen Carcinogenese an Nasenschleimhaut und Gehörgangsband der Ratte finden konnte: In den Bereichen des Epithels, die nach morphologischen Kriterien sich in ein Carcinom umzuwandeln begannen, fand Dr. Herrmann sonnenen als Prodrumalercheinung alkalische Phosphatasen in umschriebenen Epithelbezirken. Natürlich ist eine Cholesteatommatrix nicht mit einem Carcinom gleichzusetzen, aber es würde mich interessieren, ob Herr Paha zwischen dem Auftreten der Phosphatasen bei Cholesteatom und bei beginnendem Carcinom theoretisch eine Verbindung sieht.

T. Paha (Reply) to Mr Neumann. The question of Mr Neumann was much too difficult and complicated for me to offer any clear-cut answer. The only thing I should like to mention is that by the action of phosphatases much energy is being liberated. In skin, and particularly in cholesteatoma epithelium, this energy is needed in the synthesis of keratin molecules. Perhaps in early stages of the development of cancer there is an abnormal protein synthesis and consequently a great need of energy which might be provided by the action of phosphatases.

ZUR PATHOHISTOLOGIE DES DUCTUS UND SACCUS ENDOLYMPHATICUS

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Abstract. Ductus und Saccus endolymphaticus sind beim Menschen auf Grund der erhobenen histopathologischen Befunde für die Endolymphphysiologie wichtig. Sowohl die Blockade der Endolymphströmung in den Saccus, als auch Veränderungen an der Pars rugosa des Saccus führen durch Störung der Endolymphabfuhr zum Endolymphhydrops. Besonders wichtig erscheint das Verhältnis epitheliale Oberfläche zu aktivem Gefäßbindegewebe zu sein. Wegsamkeit und Zustand von Ductus und Saccus endolymphaticus sind heute von größerem klinischem Interesse als ehemals der Überleitungsweg von Infektionen.

Cotugno hat 1790 erstmals Ductus und Saccus endolymphaticus in seiner Monographie *De aquaeductibus auris humanae internae* beschrieben. Erst 1869 folgte Boetchers Darstellung und 1919 Siebenmann dessen Mitteilung, heute mit wenigen Ausnahmen Gültigkeit

finden. Silrala (1942) und Secrétan (1944) haben sich dann mit pathohistologischen Befunden auseinandergesetzt, einmal beim alten Menschen und zum anderen bei Entzündungsprozessen. Lichtmikroskopische Beobachtungen am normalen Ductus und Saccus endolymphaticus des Menschen stammen weiter von Bast & Anson (1949), Arnvig (1951), zur Pathologie von Zechner & Altmann (1969), Shambaugh (1969), Kaufmann-Ahrenberg et al. (1970).

Sicher sind Ductus und Saccus endolymphaticus kein atavistisches, der Involution anheimfallendes Organsystem. G. Portmann hatte schon 1919 und 1922 auf die Bedeutung

dieses bei der operativen Schwindelbehandlung verwiesen. Guild (1927) eine Hypothese erstellt, daß beim Hydrops endolymphaticus der Fehler in der Pars rugosa des Saccus liegen müsse. 1938 berichteten Hallpike & Cairns von der perisacculären Fibrose, Anson 1965 von der stiefmütterlichen Behandlung des perisacculären Gefäßbindegewebes. Aktivitätsnachweise, 1956 von Egmond & Brikmann gefordert, von Ductus und insbesondere Saccus endolymphaticus wurden bislang nur beim Versuchstier geführt. Im Ultramikroskop zeigt nach Lundquist et al. (1964) aber auch nach Adlington (1967) das Epithel des Saccus deutlich Merkmale des Flüssigkeitstransportes. Hohe enzymatische und damit Stoffwechselaktivität konnten Ishii et al. (1966), Schätzle & Haubrich (1966) hohen Eiweißstoffwechsel, Koburg et al. (1967) u. a. am Saccus nachweisen. Nicht unerwähnt sollten auch die Obliterationsversuche von Kimura (1967), Schuknecht et al. (1962) und Beal (1968) bleiben, welche einen Hydrops des Innenohrs erzeugen konnten bei Ausschaltung des Saccus.

Am Menschen sind wir auf Zufallsbeobachtungen angewiesen. Vergleiche zwischen Versuchstier und Mensch sind u. E. nur bedingt möglich, deshalb sind Berichte, ob von post mortem oder Operationspräparaten äußerst wertvoll. Sicher liegen Probleme vor in präparativen Schwierigkeiten. Füllnis, Fixierung, Entkalkung, aber auch in topographischer Hinsicht bzw. der Zuordnung

Herrn Univ. Prof. Dr. E. Navratil zum 70. Geburtstag in Verehrung gewidmet.

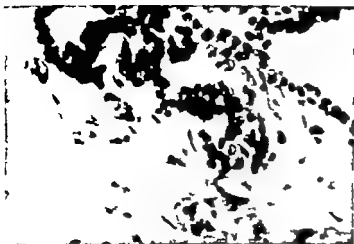


Abb. 1 Pars rugosa, Pollaks Trichrome.
Zotten mit Gefäßen im Stiel, lockeres
Bindegewebe mit weiten Saftspalten.

biopsisch gewonnener Proben. Wir glauben, daß unsere Mitteilung besonders im Hinblick auf die Beurteilung des gesamten Aquaeductus vestibuli samt Inhalt und wegen der angewendeten Präparation Aussagekraft hat.

MATERIAL UND METHODIK

Unser Material stammt ausschließlich vom Menschen, betrifft alle Altersgruppen wie schon mit Altmann zusammen teilweise 1969 mitgeteilt. Wir haben unsere Untersuchungen fortgesetzt und besonderen Wert auf die Präparation gelegt. Ausgiebige Fixierung frischer Felsenbeine in gepuffertem Formol und schonendste Entkalkung in EDTA bei pH 7,35 scheinen post mortem Artefakte weitgehend auszuschalten. Schnitten in 15–18 μ Dicke nach Parlodoneinbettung erlauben sehr gute topographische Zuordnung aller Befunde. HE-gefärbte Schnitte dienen vornehmlich diesem Zweck. Pollak's Trichromgemisch als Bindegewebsdarstellung erlaubte gute Beurteilung des Gefäßbindegewebes insbesondere in der Pars rugosa. MPS-Nachweis führten wir mit Hilfe der kombinierten Eisen-PAS-Reaktion nach Mowry Bezüglich normaler anatomischer Befunde verweisen wir auf Bast & Anson (1949) sowie eigene Veröffentlichungen. Das Fehlen einer Basalmembran haben wir 1969 am menschlichen Saccus en-

dolymphaticus mit Hilfe der Versilberung bereits widerlegen können. Neue Erkenntnisse brachten MPS-Darstellungen. Die Dokumentation unserer Befunde legen wir zum großen Teil in Farbdarstellungen dar welche auch den Wert der präparativen Technik deutlich machen.

PATHOHISTOLOGISCHE BEFUNDE

Im Vestibulum liegen die zwei Wurzeln des Ductus endolymphaticus. Ductus utriculi und sacculi vereinigen sich zum Sinus, welcher wechselnde Größe hat, ablesbar an Faltenbildungen seiner Wand. Am Beginn des Sinus springt vestibulumwärts ein Bindegewebsporn vor welcher als Bast'sche Klappe bezeichnet wird. Sie ist ähnlich der reinforced area Perlman eine widerstandsfähige Bildung, kann Verziehungen mitmachen, wird aber wie wir beim Endolymphhydrops zeigen konnten, nie aufgebraucht oder an die laterale Wand des Vestibulum gepreßt. So kann das bestehende Sinusostium nur durch überdehnte Teile des utriculus verlegt werden.

Beim Eintritt in den Aquaeductus vestibuli verengt sich der Ductus zur Pars isthmica. Hier ist er mit starken Bindegewebsfasern am Knochen befestigt. Dies ist die Stelle wo er offenbar bevorzugt postentzündlich obliteriert. Wir sahen an der Engstelle Detrituspro-

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ENDOLYMPHATICUS

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Abstract: Ductus und Saccus endolymphaticus sind beim Menschen auf Grund der erhobenen histopathologischen Befunde für die Endolymphphysiologie wichtig. Sowohl die Blockade der Endolymphströmung in den Saccus, als auch Veränderungen an der Pars rugosa des Saccus führen durch Störung der Endolymphabfuhr zum Endolymphhydrops. Besonders wichtig scheint das Verhältnis epitheliale Oberfläche zu aktivem Gefäßbindegewebe zu sein. Wegsamkeit und Zustand von Ductus und Saccus endolymphaticus sind heute von größerem klinischem Interesse als ehemals der Überleitungsraum von Infektionen.

Cotugno hat 1790 erstmals Ductus und Saccus endolymphaticus in seiner Monographie *De aquaeductibus auris humanae internae* beschrieben. Erst 1869 folgte Boetchers Darstellung und 1919 Siebenmann, dessen Mitteilung bis heute mit wenigen Ausnahmen Gültigkeit

Surala (1942) und Secrétan (1944) haben dann mit pathohistologischen Befunden auseinandergesetzt, einmal beim alten Menschen und zum anderen bei Entzündungsprozessen. Lichtmikroskopische Beobachtungen am normalen Ductus und Saccus endolymphaticus des Menschen stammen weiter von Bast & Anson (1949) Arnvig (1951) zur Pathologie von Zechner & Altmann (1969) Shambaugh (1969) Kaufmann-Ahrenberg et al. (1970)

Sicher sind Ductus und Saccus endolymphaticus kein statisches, der Involution anheimfallendes Organsystem. G Portmann hatte schon 1919 und 1922 auf die Bedeutung

dieses bei der operativen Schwindelbehandlung verwiesen. Guild (1927) eine Hypothese erstellt, daß beim Hydrops endolymphaticus der Fehler in der Pars rugosa des Saccus liegen müsse. 1938 berichteten Hallpike & Cairns von der perisacculären Fibrose, Anson 1965 von der stiefmütterlichen Behandlung des perisacculären Gefäßbindegewebes. Aktivitätsnachweise 1956 von Egmond & Brakmann gefordert, von Ductus und insbesondere Saccus endolymphaticus wurden bislang nur beim Versuchstier geführt. Im Ultramikroskop zeigt nach Lundquist et al. (1964) aber auch nach Adlington (1967) das Epithel des Saccus deutlich Merkmale des Flüssigkeitstransportes. Hohe enzymatische und damit Stoffwechselaktivität konnten Ishii et al. (1966) Schätzle & Hasubich (1966) hohen Eiweißstoffwechsel, Koburg et al. (1967) u. a. im Saccus nachweisen. Nicht unerwähnt sollten auch die Obliterationsversuche von Kimura (1967) Schuknecht et al. (1962) und Beal (1968) bleiben, welche einen Hydrops des Innenohrs erzeugen konnten bei Ausschaltung des Saccus.

Am Menschen sind wir auf Zufallsbeobachtungen angewiesen. Vergleiche zwischen Versuchstier und Mensch sind u. E. nur bedingt möglich, deshalb sind Berichte ob post mortem oder Operationspräparaten äußerst wertvoll. Sicher liegen Probleme vor in präparativen Schwierigkeiten. Füllunk, Fixierung, Entkalkung, aber auch in topographischer Hinsicht bzw der Zuordnung

Herrn Univ Prof Dr E Navratil zum 70. Geburtstag in Verehrung gewidmet.



Abb. 3 Pars rugosa. Pa-MPS-PAS Reaktion Basalmembran als Doppelkontur dargestellt zum Teil PAS positiv zum Teil tiefblau (schwarz)

narbig umgewandelt werden. Sie können auch verkleben wodurch lumenfern Zysten entstehen mit eingedicktem Inhalt oder nur Blindtasche

Auffallend gering sind die Veränderungen an der Pars intraduralis. Das schlitzförmige Lumen beinhaltet sehr oft eingedickte Massen, inwieweit dies schon krankhaft ist oder steht außerhalb unserer Erfahrungen.

Bleibt noch die Beobachtung von Makrophagen im Lumen des Sacculus (Abb 2) einmal reichlich dann wieder spärlich. Die Zellen zeigen hohe Aktivität und scheinen nicht nur corpusculäre Elemente aufzunehmen.

BESPRECHUNG UND WERTUNG DER BEFUNDE

War in der vorantibiotischen Ära das Interesse an Ductus und Sacculus endolymphaticus vorwiegend im Überleitungsweg der Infektion gelegen, so ist durch Portmann und Guild der Einfluß auf die Endolymphphysiologie in den Vordergrund getreten. Die heutige gültige Funktionstheorie des Inhalts des Aquaeductus vestibuli umfaßt Resorption und Excretion der Endolymph, Phagozytose und Abwehrmechanismus sowie Endolymphdruckregulation. Unser Bestreben ist es, die erhobenen

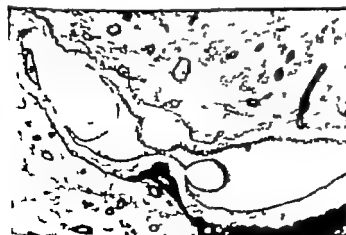


Abb. 4 Pars rugosa, IIIc. Hydrophische Zotte mit kleiner Zyste an der lumenabgewandten Wurzel, hydrophische Zotte mit metaplastischer Epithelkappe, das unter ausstülpender Sporn.



Abb. 5 Pars rugosa HE. Zotte mit inkrustiertem Inhalt als Folge von Verklebung.

pathohistologischen Befunde der Organfunktion und deren Störungen zuzunordnen.

1 Ductus und Saccus endolymphaticus haben an der Endolymphcirculation teil. Kommt es zur Unterbrechung entwickelt sich ein Endolymphhydrops. Experimentell haben es Schuknecht und Kimura mit den Obliterationsversuchen gezeigt, wir konnten es bei der bindegewebigen und knöchernen Verödung, aber auch bei Verlegung der Bast'schen Klappe sehen.

2 Aktivster Teil des Labyrinthanhanges ist die Pars rugosa des Saccus. Dies geht schon daraus hervor daß ein Hydrops entsteht, wenn die Endolymph nicht in diesen Teil gelangt. Hinweise ergeben sich auch aus dem

Bau: große epithelisierte Oberfläche, lockeres Gefäßbindegewebe, Zottenbildung und eine durchgehende Basalmembran. Diese ist reich an MPS verschiedener Art und wirkt nach Gussen sowohl als Wasser als auch als aktives Ionenfilter wegen der zahlreichen freien Endgruppen in den MPS. Zellpedikel, Varicosen, subepitheliale Vesiculation und breite Spalten sind Hinweise auf Flüssigkeitstransport.

3 Für eine Funktionsstörung der Pars rugosa scheint es morphologische Belege zu geben: sowohl hyperplastisches Bindegewebe als auch hypoplastisches und Gefäßarmut haben einen Hydrops endolymphaticus zur Folge. Es muß also um die Endolymphaufbereitung gewür-



Abb. 6 Pars rugosa. Zotten und Gefäßbindegewebsatrophie, dystrophischer Kalkherd, daneben otitische Einprägung in eine Zotte.

leistet zu sein, ein bestimmtes Verhältnis epithelialer Oberfläche zu aktivem lockerem Gefäßbindegewebe gegeben sein. Dieses Verhältnis wird grob gestört bei allen Formen der Zottenatrophie, Zottenhypophyse, Epithelmeta- und Hyperplasie sowie allen Formen der Bindegewebshyperplasie oder Hypoplasie, wenn sie mit schlechter Vaskularisation einhergeht. Unsere Ansicht bestätigen die blottischen Befunde von Shambaugh (1969) und Kaufmann-Ahrenberg et al. (1970).

4 Der Druckregelung scheinen Ductus und Saccus nur passiv und in sehr beschränktem Maße zu dienen. Zerdehnungen oder weite gebaute Lumina in der Pars intraduralis konnten wir selbst bei starkem Hydrops nicht sehen. Ob die Bast'sche Klappe eine echte Klappenfunktion erfüllt, ist nicht zu entscheiden.

5 Dem Abwehrmechanismus dienen die immer wieder zu beobachtenden Makrophagen. Diese Zellen liegen im Lumen frei und zeigen deutliche Aktivitätsmerkmale.

RÉSUMÉ

L'état morphologique des structures dans l'aqueduc du vestibule est décrit du point de vue de la fonction du ductus et du saccus endolymphatiques. L'image morphologique va de l'altération de la valve de Bast et de la partie rugueuse du saccus jusqu'à l'oblitération osseuse de l'aqueduc.

SUMMARY

Endolymphatic duct and sac are in man, involved in the endolymph physiology as documented by histopathological findings. Blockade of endolymph circulation into the sac leads, by disturbance of endolymph preparation, to an endolymphatic hydrops. The relation of epithelial surface to active vascular connective tissue seems of great importance. The message and conditions of endolymphatic duct and sac are nowadays of more clinical interest, than in the past as a way of infection.

LITERATUR

Adlington, P. 1967 The ultrastructure and the function of the saccus endolymphaticus and its decompensation in Ménière's disease. *J Laryng* 81 759

- Altman, F. & Zechner, G. 1968 The pathology and pathogenesis of endolymphatic hydrops. *Arch Klin Exp Ohr Nas Kehlkopfheilk* 192 1.
- Anson, B. J. 1963 The endolymphatic and perilymphatic aqueducts of the human ear. *Acta Otolaryng* (Stockh.) 59 140.
- Arnvig, J. 1951 Lymphocytes in the wall of the endolymphatic sac. *Arch Otolaryng* (Chic.) 53 290.
- Bast, T. H. & Anson, J. B. 1949 *The temporal bone and the ear*. Thomas, Springfield, Ill.
- Beal, D. D. 1968. Effect of endolymphatic sac ablation in the rabbit and cat. *Acta Otolaryng* (Stockh.) 66 333.
- Borchert, A. 1869 Über den Aquaeductus vestibuli bei Katzen und Menschen. *Arch Anat Physiol* 56 37.
- Egmond, A. A. J. van & Brinkmann, W. F. B. 1956. On the function of the saccus endolymphaticus. *Acta Otolaryng* (Stockh.) 46 283.
- Guild, S. R. 1977 The circulation of endolymph. *Amer J Anat* 59 57.
- Gussen, R. 1966. Vesicles and basement membrane changes associated with hydrops of the saccule and endolymphatic duct and sac. *Acta Otolaryng* (Stockh.) 62 403.
- 1966. Basement membranes in the ear. *Ann Otol* 75 1124.
- Hallpike, C. E. & Cairns, H. 1938. Observation on the pathology of Ménière's syndrome. *J Laryng* 53 625.
- Ishii, T., Silverstein, H. & Balogh, K., Jr. 1966. Metabolic activities of the endolymphatic sac. *Acta Otolaryng* (Stockh.) 62 61.
- Kaufmann-Ahrenberg, J., Marowitz, W. F., Shambaugh, G. E. 1970. The role of the endolymphatic sac in the pathogenesis of endolymphatic hydrops in man. *Acta Otolaryng* (Stockh.), Suppl. 275.
- Kimura, R. & 1967 Experimental blockage of the endolymphatic duct and sac and its effect on the inner ear of the guinea pig. *Pract Otorhinolaryng* (Basel) 76 665.
- 1967 Experimental production of endolymphatic hydrops. *Int Symp Meniere's Dis.* Mayo Clinic, Rochester Minn.
- Kimura, R. S., Lundquist, F. G. & Wersäll, J. 1964. Secretory epithelial cells in the ampullae of the guinea pig labyrinth. *Acta Otolaryng* (Stockh.) 57 517.
- Kimura, R. S. & Schuknecht, H. F. 1965 Membranous hydrops in the inner ear of the guinea pig after obliteration of the endolymphatic sac. *Pract Otorhinolaryng* (Basel) 27 343.
- Koburg, E., Haubrich, J. & Kernbach, B. 1967 Autoradiographische Untersuchungen zum Stoffwechsel des Ductus und Saccus endolymphaticus. *Acta Otolaryng* (Stockh.) 64 146.
- Lundquist, F. G., Kimura, R. S. & Wersäll, J. 1964. Ultrastructural organization of the epithelial lining in the endolymphatic duct and sac of the guinea pig. *Acta Otolaryng* (Stockh.) 57 65.
- Portmann, G. 1919 Recherches sur le sac et le canal endolymphatiques. *C.R. Soc Biol (Par)* 82 1384.

- 1920. *Ibid.* 83 45
- 1921 *Ibid.* 85 1070.
- Schlöde W & Haubrich, J 1966. Über Verteilung und Glycosidasen, Esterasen und Eiweißhaustellen im Saccus endolymphaticus des Meerschweinchens. *Arch Ohr Nas Kehlkopfheilk* 186 373
- Schuknecht H F, Benitez, J T & Berkhuys, J 1962. Further observations on the pathology of Menière disease. *Ann Otol* 71 1039
- Secretan, J P 1944. De l'histologie normale du sac endolymphatique chez l'homme. *Acta Otolaryng* (Stockh.) 32 119
- 1944. Der Saccus endolymphaticus bei Entzündungsprozessen. *Pract Otorhinolaryng* (Basel) 6 1
- Shambaugh G E. 1969. Observations on the endolymphatic sac in cases of hydrops. *Arch Otolaryng* (Chic.) 89 98
- Siebenmann, F 1919. Anatomische Untersuchungen über den Saccus u. Ductus endolymphaticus beim Menschen. *Pawlow Schäfer Beitr Anat Ohr* 13 59
- Silrala, U 1942. Über den Bau und die Funktion des Ductus und Saccus endolymphaticus beim alten Menschen. *Z Anat Entwicklungsgesch* 11 246.
- Silverstein, H. 1966. Biochemical and physiological studies of the endolymphatic sac in the cat. *Laryngoscope* 76 498.
- Zechner G 1971. Die Pathohistomorphologie des endolymphatischen Hydrops. *Jena-DDR Symposium für Cochleaforschung* Im Druck.
- 1971. Morphologische Beobachtungen beim Innenohrhydrops. *Mtschr Ohrenheilk*. Im Druck.
- 1972. Pathohistologie des blutigen Labyrinthas beim endolymphatischen Hydrops. *Menière-Symposium Wien. Mtschr Ohrenheilk*. Im Druck.
- Zechner G & Altmann, F 1969. Histological studies on the human endolymphatic duct and sac. *Pract Otorhinolaryng* (Basel) 31 63

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DISKUSSION

K H Fosterm. Kennen Sie Fälle mit endolymphatischem Hydrops ohne Veränderungen im Saccus? Und kennen Sie Fälle mit verschlossenen Ductus oder Saccus ohne Hydrops? Ich habe solche Präparate in der Wittmaas'schen histologischen Sammlung gesehen.

U Fisch. I would like to ask Mr Zechner if he could say something about the frequency of the changes he has observed. At surgery we found a total obliteration of the endolymphatic sac in 20% of the cases and a partial obliteration in 30%. We are faced with 30% macroscopically "normal" looking sac in Menière's disease.

G Zechner (Reply) to Mr Fosterm. In agreement with earlier findings we never saw endolymphatic hydrops without alterations of the endolymphatic duct and sac; this is also confirmed by Shambaugh (lack of vascularization). Kaufmann-Ahrenberg, Marink and Shambaugh (histologically proven, lack of vascularization) Sabotik showed recently in a Mondini-type of hearing loss an aplasia of ductus endolymphaticus and bulging Reissner's membrane as a sign of endolymphatic hydrops.

To Mr Fisch. In all our hydrops cases we saw various changes microscopically in the vestibular aqueduct. Pure macroscopic findings alone are not so significant for function of the system, although Shambaugh found poor vascularization in operations on the sac.

DIE HEMMUNG VON LEUKOZYTEN — UND GEWEBSKATHEPSINEN DURCH DEN INHIBITOR DES FLIMMEREPITHELS UND DAS ALPHA 1 ANTITRYPSIN DES SERUMS

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Abstract: Proteasen aus zerfallenen Leukozyten zerstören körpereigenes Eiweiß, wenn sie nicht durch das alpha₁-Antitrypsin des Serums gehemmt werden. Das Flimmerepithel produziert einen niedermolekularen, saurestabilen Proteaseninhibitor (n.a.i.), der eine ausreichend autolytische Konzentration im Nasen- und Bronchalsekret aufrecht erhält. Bei akuten und chronischen Infektionen ist diese Inhibitor-Konzentration erniedrigt, bei der Ozaena aufgehoben. Leukozytäre Proteasen zeigen zwei verschiedene Wirkungen: (1) die Destruktion von Basalmembranen und (2) die Aktivierung des Kallikrein-Kinin-Systems, das die Kardinalsymptome der Entzündung auslöst. Die Isolierung von Proteasen aus Eiter ergibt 5 aktive Komponenten, wobei die Fraktion mit der Hauptaktivität durch den n.a.i. voll gehemmt ist. Die Simultanelektrophorese des Serums bei schweren eitrigen Nebenhöhleninfektionen zeigt hinter dem alpha₁-Antitrypsin-peak einen zweiten peak, der den Proteasen-alpha₁-Antitrypsin-Komplex darstellt.

Leukozyten interessieren den Kliniker solange sie als intakte Zellen zu zählen und zu färben sind. Das Interesse sinkt, wenn Leukozyten als Eiter auftreten und erlischt mit dem Zerfall dieser Zellen. Die Erkenntnis, daß Leukozyten auch nach ihrem Zerfall von großer klinischer Bedeutung sind, geht auf die Beobachtung zurück, daß bei noch jugendlichen Patienten mit schweren Lungenemphysemen eine bestimmte elektrophoretische Fraktion im Serum, nämlich die alpha₁-Globulin-Fraktion, fehlt (Laurell & Eriksson, 1963). Aus dem Nachweis der Hemmbarkeit aus Leukozyten stammender Proteasen durch das in der alpha₁-Fraktion wandernde Globulin, das als alpha₁-Antitrypsin bekannt ist, ergab sich die

Erklärung für die jugendlichen Emphyseme beim Fehlen der alpha₁-Fraktion (Liebermann & Gawad 1971, Ohlson, 1971). In derartigen Fällen werden nämlich die in der Lunge vermehrt anfallenden Leukozytenproteasen nicht ausreichend gehemmt und spalten körpereigene Strukturen wie z. B. Basalmembranen (Frimmer 1971). Ähnliche physiologische Hemmmechanismen als Schutz des Organismus vor Autolyse wurden zuerst am Pankreas beobachtet und sind z. B. auch von der Blutgerinnung her bekannt. Wir konnten nachweisen, daß ein solcher Vorgang auch im Bereich des Flimmerepithels, also im Bereich der Atemwege von Bedeutung ist. Wir wiesen im Nasen- und Bronchalsekret einen niedermolekularen Proteaseninhibitor nach, der eine Sekretionsleistung des Flimmerepithels darstellt (Hochstrasser et al., 1971 und 1972). Auch dieser Inhibitor hemmt leukozytäre Proteasen, die im Verlauf von Infekten der Atemwege durch Leukozytenemigration — und Zerfall massiert frei werden. In den Sekreten Gesunder liegt dieser Inhibitor in bestimmten Konzentrationen vor. Er ist erniedrigt bei Infekten der Atemwege durch verminderte Produktion und durch Bindung an leukozytäre Proteasen. Er fehlt bei schweren eitrigen Infekten und bei der Ozaena (Reichert et al., 1971, Reichert & Hochstrasser 1972).

Der Inhibitor schützt das Flimmerepithel also vor der direkten proteolytischen Einwirkung

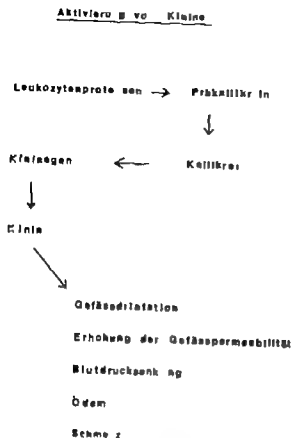
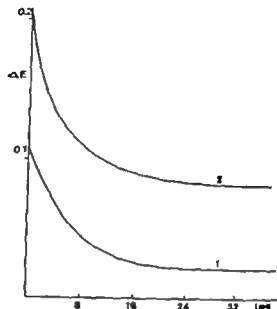


Abb 1

kung leukozytärer Proteasen. Gleichzeitig wird in der Anwesenheit des Inhibitors noch ein anderer Vorgang unterdrückt, der durch leukozytäre Proteasen ausgelöst werden kann, die Freisetzung von Kininen.

Proteasen aus Leukozyten können wie Entzündungsmediatoren wirken und das Kallikrein-Kininsystem aktivieren. Sie können das ubiquitär vorkommende Präkallikrein durch Proteolyse in aktives Kallikrein umwandeln. Dieses Kallikrein aktiviert seinerseits Kininogen zu (Abb. 1) kinin, einer hochaktiven pharmakologischen Substanz. Kinine lösen die Kardinalsymptome der Entzündung aus. Sie senken den Blutdruck durch Gefäßdilatation erhöhen die Gefäßpermeabilität stören die Mikrozirkulation durch Ödembildung und lösen Schmerz aus. Die Beobachtung, daß Leukozytenproteasen, die sowohl autolytisch wie kininfreisetzend wirken durch den Inhibitor des Flimmerepithels hemmbar sind,



Hemmung von Leukozytenprotease durch

1 α_1 Antitrypsin

2 Flammoeptithel

Abb 2

und die Absicht diese Beziehung eines Tages klinisch-therapeutisch nutzen zu können, veranlaßte uns, diese Enzyme genauer zu untersuchen.

In unseren ersten Untersuchungen, in denen wir die Hemmbarkeit der Proteasen durch den Inhibitor nachweisen verwendeten wir Elter aus Kieferhöhlen der ja ein Gemisch verschiedenster Proteasen darstellt. Wir ließen den Elter auf natürliche Substrate wie Azocasein und Hämoglobin einwirken, setzten in Parallelansätzen Inhibitor hinzu und stellten fest, daß die proteolytische Gesamtaktivität (Abb. 2) von dem niedermolekularen Inhibitor zu 80% und von dem α_1 -Antitrypsin des Serums zu 60% gehemmt wird. Die nicht hemmbare Restaktivität von 20 bzw 40% führten wir auf Anteile nicht hemmbarer Exopeptidasen zurück. Zur genaueren Differenzierung versuchten wir das Proteasengemisch chromatographisch aufzutrennen und geson-

dert auf ihre Hemmbarkelt zu untersuchen. Durch Chromatographie an DEAE Sephadex gelang eine Auftrennung in 7 Komponenten. Komponente IV und VII enthält keine nennenswerte proteolytische Aktivität. Die aktiven anderen Komponenten wurden an Sephadex G-100 weiterfraktioniert. Dabei wurden letztlich 5 Komponenten gewonnen, die die Hauptaktivität enthalten (Abb 3). Von den verschiedenen Komponenten ist nur die Fraktion V₄, die allerdings die Hauptaktivität enthält, voll hemmbar und zwar sowohl durch unseren Inhibitor wie durch alpha₁-Antitrypsin. Dem chemischen Verhalten nach handelt es sich dabei um die Protease, die Basalmembranen abbauen kann.

Nachdem es uns gelungen war aus Leukozyten isolierte Proteasen mit unserem Inhibitor zu hemmen, interessierte es uns zu prüfen, ob auch andere lysosomale Proteasen durch diesen Inhibitor zu hemmen sind. Wir wählten Gewebsproteasen aus der Milz und stellten fest, daß die proteolytische Wirkung solcher Lysosomenpräparate durch unseren Inhibitor nur zu 30% hemmbar ist. Diese Ergebnisse belegen die Hemmspezifität des niedermolekularen Inhibitors und bestätigen damit seine Funktion als Hemmstoff leukozytärer Proteasen im Nasen- und Bronchalsekret.

Nachdem wir das Verhalten unseres Inhibitors in den Sekreten weitgehend geklärt hatten beschäftigten wir uns mit dem Verhalten des alpha₁-Antitrypsins, des erwähnten Proteasehemmstoffs im Serum, der in geringer Konzentration auch in den Sekreten der Atemwege erscheint. Dabei stellten wir in der Immunoelektrophorese eitriger Bronchalsekrete fest, daß hinter der alpha₁ Fraktion ein zweiter Peak erscheint, den wir als den verzögert wandernden Komplex von alpha₁-Antitrypsin mit leukozytären Proteasen auffaßten. In vitro ließ sich dieser Vorgang nachvollziehen, indem wir Serum mit Leukozytenproteasen versetzten und im elektrischen Feld wandern ließen. Mittlerweile gelang es, diesen Komplex u. a. auch in eitrigen Liquor nachzuweisen. Bei schweren lokalen Eiterungen in unserem

Spezifische Aktivität
von
Leukozytenproteasen
gegen
Hämoglobin

Δ E/mg Protein/h

L I 0 251

L II 0 075

L V₁ 0 056

L V₃ 3 44

L VI₁ 0.071

Abb 3

Fachgebiet, Stirnbeinosteomyelitis, Orbitalphlegmone u. a. beobachteten wir einerseits im Sekret aufgehobener Konzentration an niedermolekularem Inhibitor und andererseits ein deutliches Vorkommen dieses Proteasen-alpha₁-Antitrypsinkomplexes im Serum.

Bis jetzt ist nicht geklärt, ob das z. B. bei chronisch rezidivierenden Schleimhauterkrankungen nachgewiesene Inhibitordefizit Folge jener Schleimhautaffektionen oder als Ausdruck einer konstitutionellen Produktionsschwäche Ursache jener erhöhten Erkrankungsrate ist. Angesichts der nachgewiesenen Bedeutung eines ausreichenden Inhibitorspiegels für die Inaktivierung der Leukozytenproteasen erhebt sich darüberhinaus die Frage, ob und auf welche Weise bei Inhibitor-mangel durch Inhibitorsubstitution ein therapeutischer Effekt erzielt werden kann.

RESUME

Les cathepsins sont des enzymes lysosomales et agissent comme médiateurs d'inflammation. Elle de-

libèrent de l'histamine et de la kénine, fortifient la perméabilité vasculaire, empêchent la microcirculation et causent la douleur et l'œdème. Quelques unes d'elles détruisent même les membranes basales. L'inhibiteur de l'épithélium respiratoire, ainsi que l'alpha-antitrypsin inhibent certaines de ces cathepsines. C'est pourquoi ces inhibiteurs influencent le processus d'inflammation.

SUMMARY

Proteases derived from disintegrated leukocytes will split protein structures of the human organism if they are not inhibited by the alpha-antitrypsin of blood-serum.

The respiratory epithelium produces its own low molecular inhibitor (Lm.i.) to maintain an adequate antiproteolytic concentration in nasal and bronchial mucus. This Lm.i. concentration is reduced during acute and chronic infections and is absent in ozema.

Leukocyte proteases exhibit two different functions: (1) destruction of protein elements such as basal membranes; (2) activation of the kallikrein-kinin-system which leads to the cardinal symptoms of inflammation.

The isolation of proteases from pus reveals 5 active compounds with the main proteolytic activity in a certain fraction, which is completely inhibited by the low inhibitor.

The crossed serum immunoelectrophoresis of patients with severe paranasal inflammation reveals behind the alpha-antitrypsin a second peak representing the complex of alpha-antitrypsin and proteases.

LITERATUR

- Friemer M. 1971 Biochemie und Pathophysiologie von Entzündungsmediatoren. *Int Z Klin Pharm Ther Toxicol* 2 144
- Hochstrasser K., Haendle H., Reichert, R. & Werle, E. 1971 Über Vorkommen und Eigenschaften eines Proteaseninhibitors im menschlichen Nasensekret. *Z Physiol Chem* 352 954
- Hochstrasser K., Reichert, R., Schwarz, S. & Werle, E. 1971. Isolierung und Charakterisierung eines Proteaseninhibitors aus menschlichen Bronchialsekret. *Z Physiol Chem* 353 im Druck.
- Laurell, C. B. & Eriksson, S. 1963 The electrophoretic alpha-globulin pattern of serum in alpha-antitrypsin deficiency. *Scand J Lab Invest* 15 132.
- Liebermann, J. & Gawad, M. A. 1971 Inhibitors and activators of leucocytic proteases in purulent sputum. *J Lab Clin Med* 77 713
- Ohlson, K. 1971 Neutral leucocyte protease and elastase inhibited by plasma alpha-antitrypsin. *Clin Lab Invest* 28 251
- Reichert, R. & Hochstrasser K. 1972 Veränderungen des Proteaseninhibitorspiegels im menschlichen Nasensekret im Verlauf verschiedener Rhinopathien. *Z Laryng Rhinol Otol* 51 73
- 1972. Proteaseninhibitormangel im Nasensekret des Ozemakranken. *Arch Klin Exp Ohr Nas Kehlkopfheilk* 201 11
- Reichert, R., Hochstrasser K. & Werle, E. 1971 Der Proteaseninhibitorspiegel im menschlichen Nasensekret unter physiologischen und pathophysiologischen Bedingungen. *Klin Wochr* 49 1234
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DISKUSSION

Mr Arsten, fragt, ob die interessanten Ergebnisse der Nasenschleimhaut-Biochemie Schule von Prof. Kammann Aufklärungen über die Immunitäts-Mechanismen geben können, die heute eine große Rolle in der Ätiologie der Entzündungen der chronischen Krankheiten der Nasenschleimhaut spielen. Ein bedeutendes Beispiel dieser Pathologie ist die Ozema, die in der HNO-Klinik der Universität von Padua als autoimmunologische Krankheit studiert worden ist.

R. Reichert (Antwort) zu Mr Arsten. Die Anwesenheit des niedermolekularen Proteaseninhibitors im Nasensekret ist unabhängig von immunologischen Vorgängen an der Schleimhaut. Jedenfalls fanden wir in den bisher untersuchten Sekreten von allergischen Rhinitiden weitgehend normale Inhibitorgehalte. Unabhängig von immunologischen Momenten halten wir es für durchaus möglich, daß das Fehlen von Inhibitor im Sekret des Ozemakranken ätiologische Bedeutung haben kann.

ZUR ENDOSKOPISCHEN ANATOMIE DER MENSCHLICHEN
SIEBBEINMUSCHELN

W. Messerklinger

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Abstrakt. Die Siebbeinmuskeln können bei der Nasenendoskopie eingehend untersucht werden. Damit werden bei ihren vielen anatomischen Variationen, die sich aus der Ethmoidentwicklung ableiten, auch für klinische Belange interessant: die Zelle im Muschelkopf der Muschelsinus, die Porens oder Incisur im 1. Ethmoidsinus, die Muschelzahl, die Teilungsbildung und die Muschelrudimente. Pathologische Veränderungen können nur mit Kenntnis der anatomischen Variationen einwandfrei identifiziert werden; sie reichen von der Taile am freien Muschelrand bei einer kranken Muschelzelle über den Schleimhautprolaps aus dem Muschelstumpf mit Furchenbildung am freien Muschelrand bis zur Schleimhaut-Schwellung bzw. Proliferation an der oberen Muschel bei chronischem Hirndruck.

Für die Routine der täglichen rhinologischen Praxis war an den Siebbeinmuskeln bis heute zu Besonderheiten lediglich die selten diagnostizierte Concha bullosa von Interesse, während die Vielzahl der anatomischen Varianten, da mit den herkömmlichen Untersuchungsmethoden nicht sichtbar für die Klinik keine Bedeutung erlangt haben und so praktisch vergessen wurden. Die Nasenendoskopie (Messerklinger 1970) ermöglicht jetzt aber auch einen weiten Einblick in das Ethmoid, damit ist es notwendig, sich mit den anatomischen Variationen des Siebbeinlabyrinths vertraut zu machen, um sie von Erkrankungen differenzieren zu können. Hier sollen nur jene Gebilde besprochen werden, die nach der Definition von Gegenbaur (1880) „von der Wand her entspringende selbständige, von einer einfachen Fortsetzung des Skeletts der

Wand gestützte Einragungen sind die Bulla ethmoidalis und der Processus uncinatus, die entwicklungsgeschichtlich Zwischenmuskeln sind (Peter 1925 Starck, 1965) seien hier ausgeklammert.

Die untere Siebbeinmuschel bildet in ihrem mittleren und hinteren Drittel durch Einrollen ihres freien Randes nach lateral den Muschelsinus, in den wiederholt die Bulla ethmoidalis ragt. Durch zarte Knochenbrücken wird der Sinus nicht selten in einzelne Nischen unterteilt und kann sich nach vorne blasig ausweiten. Diese Pneumatisation im Muschelkopf, die schon beim Neugeborenen beginnt (Richter 1932) und extrem ausgebildet als Concha bullosa bezeichnet wird, mündet bei kompletter knöcherner Umgrenzung durch ein Ostium an der lateralen Muschelfläche in Höhe des Kieferhöhlenostiums, während sie andernfalls breit in den Muschelsinus übergeht und durch ihn breit mit dem mittleren Nasengang kommuniziert.

Der Muschelkopf scheint dann eine Zelle zu enthalten, wenn er abgerundet ist, während er andernfalls so schmal wie die übrige Muschel ist. Erkrankt eine Muschelzelle, dann schwillt die Schleimhaut über dem Muschelkopf an am Übergang zum gesunden Muschelteil grenzt sich die Mucosa am freien Rand durch eine typische Taille ab. Dauert die Entzündung längere Zeit kann am Muschelkopf die Schleimhaut warzig hyperplastisch und knapp medial von der Kante des Muschelhalbes ein solitärer Polyp entspringen,

Herrn Prof. Dr. Robert Stigler zum 95. Geburtstag gewidmet.

der sich im Fornix bis in den Introtitus nasi erstrecken kann. Bricht eine kranke Zelle nach vorne durch, so entsteht zuerst am Muschelkopf ein nekrotischer Belag und schließlich kann sich ein solitärer Polyp aus der Zelle stülpen, dessen Stiel auffällig gut zu sehen ist. Nach Abtragen des Polypen findet sich ein nach vorne offener von zarten Knochenwänden umgebener Trichter der mit oedematös-polypöser Schleimhaut ausgekleidet ist. Aus dem Ostium einer kranken Muschelzelle im mittleren Nasengang kann pathologisches Sekret abfließen oder die Mucosa sich halbkugelig vorstülpen.

Auch der Muschelsinus kann vom mittleren Nasengang aus mit Winkelspektiven eingesehen werden. Erkrankt er dann wölbt sich die sehr schwellungsfähige Siebbeinschleimhaut oedematös-entzündlich, hyperplastisch oder polypös verändert in den mittleren Nasengang vor und kann ihn ausfüllen. Am freien Muschelrand entsteht dabei am Übergang zur weniger schwellungsfähigen Mucosa der Muscheloberfläche eine seichte Furche. Lateral der Schwellung ist der mittlere Nasengang freisondierbar. Ähnliche Erscheinungen, wie z.B. ein überlappendes mit hyperplastischer Schleimhaut überzogener Processus uncinatus, kranke Agger nasi-Zellen oder ein Prolaps einer vorderen unteren Fontanelle sind durch leicht vom kranken Muschelsinus zu differenzieren (Abb. 1 10-20 bei Messerklinger (3)).

In den meisten Fällen kann man mit dem Endoskop über den Recessus sphenoethmoidalis, manchmal auch zwischen Septum und mittlerer Muschel tief in das Siebbein eindringen und die weiteren Muscheln und Nasengänge mit ihren verschiedenen Varianten nebst Furchen- und Spaltbildung untersuchen. Die Vielfalt dieser Bildungen ist nur aus der Kenntnis der Ethmoidentwicklung zu verstehen. Es bestehen zwar in dieser Frage noch verschiedene Meinungen, die vor allem dann hervortreten, wenn man z.B. die Artikel von Hochstetter (1944), Killian (1896), Grünwald (1917) Peter (1913) und Richter (1932) ne-

beneinander stellt, das für hier Wichtiges dürfte aber im Wesentlichen geklärt sein.

Durch Eindringen von Epithelfurchen oder Rinnen und Gewebsvermehrung der so herausgeschnittenen Wülste werden die Ethmoidmuscheln gebildet, wobei die Epithelfurchen primär gestaltend sind (Grünwald, 1917). Von den Hauptwülsten-Hauptmuscheln werden nach Killian 5-6, nach Peter höchstens 3, Richter 3 und Grünwald je nach Individuum 3 oder 2 angelegt, wobei in der Reife der größere Teil der 2-wulstigen durch Reduktion aus 3-wulstigen entsteht. 23% von entwickelten plus 10% rudimentären obersten Muschel beim Erwachsenen stehen 48% im 8.-10. Foetalmonat angelegten gegenüber (Grünwald). Peter will hingegen in der Reife nur 2 Hauptmuscheln ausgebildet sehen und klassifiziert eine oberste Muschel als Tellmuschel der oberen. Dem hält Richter entgegen, daß in der obersten Muschel später als in der oberen die Knochenbildung einsetzt und eine Concha suprema wiederholt größer ist als eine obere Muschel. Vor allem auf dem 1. Ethmoidnase kommen Nebenfurchen vor die eine mehr oder weniger ausgeprägte Teilwulstbildung bedingen können. Im 5. Foetalmonat fand sie Grünwald in 59% beim Erwachsenen in 6%. In seltenen Fällen können die Furchen und Spalten die hintere oder vordere Grenz der Muschel überschreiten.

Endoskopisch konnte ich am Menschen beobachten, meist ist nur eine obere Muschel vorhanden, die frei endet (Abb. 10 bei Messerklinger (2)) oder auch in die Rückwand des Recessus sphenoethmoidalis auslaufen kann; ihr vorderes Ende kann schlank sein oder auch einen Lobulus aufweisen. Im darunter liegenden Nasengang kann sich mitunter eine schmale Leiste finden, die nicht mehr das Niveau der Muschel erreicht und auch nicht mehr die ganze Länge des Meatus einnehmen muß. Sie ist eine rudimentäre 2. Siebbeinmuschel, während das anfangs als obere Muschel imponierende Gebilde eine Concha suprema ist (Abb. 11 bei Messerklinger (2)). Diese Bildungen lassen sich zwanglos in die

Beobachtungen Zuckerkandls (1893) oder die verschiedenen Typen Grünwalds emreihen. Eine 2. und 3. Siebbeinmuschel kann aber auch voll ausgebildet sein und die 3. sogar ein Tuberculum ethmoidale posticum (Zuckerkandl) aufweisen, das in den obersten Nasengang mündet (Abb. 1). Auch ein 4. Ethmoturbinale kann in seltenen Fällen vorkommen (Abb. 2) ob es ein Teilwulst oder eine Hauptmuschel ist, läßt sich aus der Beobachtung allein nicht entscheiden.

Furchen verschiedener Länge finden sich mit Vorliebe an der medialen Fläche des 1. Ethmoturbinales, knapp über dem freien Rand und mit ihm parallel laufend (Abb. 2 bei Messerklinger (3)). Schneiden sie ausnahmsweise tiefer ein, so können sie eine mehr oder weniger ausgeprägte Teilwulstbildung bedingen. Diese Teilmuskeln können ein gemeinsames Ende haben, oder was selten ist, ihr Ende durch eine Incisur geteilt und damit schwalbenschwanzartig sein (Abb. 3 bei Messerklinger (3)). Schwieriger wird die Entscheidung, wenn knapp hinter dem Kopf der 1. Siebbeinmuschel und manchmal auch noch unter ihrem Hals eine Muschel durch eine tiefe Furche abgetrennt ist (Abb. 3), die fallweise im vorderen Abschnitt bis an den freien Rand der 1. Siebbeinmuschel herabreichen (Abb. 4) und damit große Ähnlichkeit mit dem 2. Ethmoturbinale des Schimpansen haben kann (Zuckerkandl). Der freie Rand eines solchen Gebildes verläuft nach rückwärts aufwärts und endet, wiederholt in die Rückwand des Recessus sphenoeithmoidalis übergehend in derselben Höhe wie eine 2. Siebbeinmuschel, gerade der letztere Grund könnte dafür sprechen, daß es sich bei dieser Bildung doch um eine 2. Siebbeinmuschel handelt und nicht um eine Teilmuschel des 1. Ethmoturbinales. Ebenfalls selten kann zwischen dem Hals der 1. Siebbeinmuschel und dem Septum der Kopf eines Ethmoturbinales vorragen und in der Frontalebene bis oder über den Kopf der mittleren Muschel reichen (Abb. 5).

Finden die bisherigen Beobachtungen noch mit den vielfältigen Möglichkeiten aus der

Siebbeinentwicklung eine Erklärung, so komplizieren Doppelungen des Kopfes der mittleren Muschel, und zwar Doppelungen zwischen Septum und Muschelkopf und nicht wie aus der Pathologie bekannt im mittleren Nasengang das Bild außerordentlich. Ich konnte bisher zwei solche Fälle beobachten. Im ersten war die mediale Doppelung durch eine Furche vom Muschelkopf getrennt nach dem Tastbefund fehlte ihr eine knöcherne Grundlage und als man mit dem Endoskop in die Furche eindrang, erschien dahinter eine Muschel, die mit der mittleren gemeinsam endete. Im zweiten Fall fand sich ein zu großer weicher Muschelkopf an dessen medialer Fläche eine breite Furche begann (Abb. 6, 7), die bis knapp vor das hintere Ende der Concha media in zwei Teilmuskeln trennte (Abb. 8) ungefähr am Ende des Muschelkopfes strahlte eine zweite frontal verlaufende Furche in die erstgenannte sagittale ein und setzte so den fleischigen Kopf deutlich von der Muschel ab (Abb. 9). In der anderen Nasenhälfte desselben Individuums war der Kopf der mittleren Muschel ebenfalls größer und hatte an der medialen Fläche ein tiefes Grübchen (Abb. 10). Diese Bildung des Muschelkopfes aus der Siebbeinentwicklung zu erklären ist unmöglich. Rächter & Spuler kamen bei ihren entwicklungsgeschichtlichen und anatomischen Studien zur Überzeugung, daß ein Rest des Nasoturbinales im nach abwärts gerichteten vorderen Ende der mittleren Muschel enthalten ist. Dies könnte auch die zwei angeführten Beobachtungen erklären und wird auch dadurch wahrscheinlich, als ich bei einem meiner Fälle an der lateralen Fläche einer mittleren Muschel am Ende des Muschelkopfes eine fast vertikal verlaufende Furche beobachten konnte (Abb. 11), die ebenfalls aus der Siebbeinentwicklung nicht erklärbar ist.

Furchen am freien Rand des 2. oder 3. Ethmoturbinales sind bisher nicht beobachtet worden. Am freien Rand einer oberen Muschel konnte ich eine Furche beobachten, die beidseits von etwas verdickter oder gestauter

Schleimhaut begrenzt war und eine Ähnlichkeit mit der Furche am freien Muschelrand bei einer Erkrankung des Muschelnus hatte (Abb. 12). Es fehlte jeder Anhalt für eine entzündliche Genese. Da die Schwellung im hinteren oberen Recessus sphenothmoidalis zunahm und den Blick zum Keilbeinostium verlegte, wurde der 56 Jahre alte Kranke eingehend untersucht und dabei ein chronischer Hirndruck mit Stauungspapille als Folge eines großen parasagittalen Meningeoms gefunden. Drei Monate nach der Operation war die Furche noch zu sehen, die Schleimhaut aber unauffällig und der Blick zum Ostium sphenoidale frei. Die Schleimhautschwellung dürfte somit die Folge des Hirndrucks gewesen sein. Beim Tier sind ja innige Verbindungen zwischen dem Subarachnoidalraum und dem Lymphgefäßnetz der Nasenschleimhaut bekannt, die nach den Untersuchungen von Hoffmann & Thiel (1956) wahrscheinlich über die Perineuralscheiden des N. olfactorius und die Gewebespalten in die Nasenlymphgefäße gehen. Beim Menschen konnte eine solche Verbindung bisher nur im 1. Lebensjahr nachgewiesen werden (André, 1905; Zwilling, 1914), während sie beim Erwachsenen ganz oder größtenteils obliteriert sein soll (Grünwald). Daß die oben angeführte Beob-

achtung kein Zufall ist, zeigte mir die Untersuchung eines weiteren chronischen Hirndrucks bei einem 5 Jahre alten Kind, das an einem Kleinhirntumor erkrankt war: die Mucosa war zwar nicht wesentlich gestaut und auch der Blick zum Ostium sphenoidale war frei, die Reflexe auf der Schleimhaut der oberen Muschel und des hinteren oberen Septums zeigten aber eine deutliche Pulsation.

Die Folgerung aus diesen Beobachtungen ist, daß nur die Kenntnis der vielen aus der Siebbeinentwicklung möglichen anatomischen Variationen es ermöglicht, endoskopisch pathologische Veränderungen an den Eihmurturbinalen zu erkennen und einwandfrei zu identifizieren.

RÉSUMÉ

Il est possible d'examiner à fond les cornets de l'ethmoïde grâce à l'endoscopie endonasale. Ainsi ses multiples variations anatomiques dérivant du développement de l'ethmoïde sont également intéressantes pour le domaine clinique: la cellule dans la tête du cornet, le sinus ethmoïdal, le sillon ou l'incision du premier cornet de l'ethmoïde, le nombre de cellules ethmoïdales, la formation partielle des cellules et les ébauches de l'ethmoïde. Les malformations pathologiques sont entièrement identifiables seulement quand on connaît les variations anatomiques; elles vont de l'entaille au bord libre de l'ethmoïde dans le cas d'une cellule ethmoïdale malade en passant par le prolapsus de la muqueuse provenant du sinus

Abb. 1 Linke Nase: Eihmurturbinale II und III mit Tuberculum ethmoidale posticum und Mündung einer Siebbeinzelle in den obersten Nasengang. Abkürzungen: Am = Agger nasi ET I, II, III = Eihmurturbinale ET I, II, III F = Foveola I = Incisur S = Septum Tep = Tuberculum ethmoidale posticum.

Abb. 2 Rechte Nase: Eihmurturbinale II, III und IV

Abb. 3 Linke Nase: von der medialen Fläche des Eihmurturbinales I entspringende Concha, die wie ein Eihmurturbinale II endet

Abb. 4 Linke Nase: von der medialen Fläche des Eihmurturbinales I entspringende Concha, die nach caudal bis an den freien Rand der mittleren Muschel reicht und rückwärts wie ein Eihmurturbinale II endet.

Abb. 5 Linke Nase: knochenartige Hyperplasie des Kopfes von Eihmurturbinale II die in der Frontalebene weit den Hals von Eihmurturbinale I nach vorne überragt.

Abb. 6 Linke Nase: lappige Hyperplasie des Kopfes von Eihmurturbinale I mit halbkehlartiger Furchen-

bildung an der medialen Seite des Kopfes, die bis knapp vor das Muschelende reicht.

Abb. 7 Wie Abbildung 6: Blick in die Furchen durch eine zusätzliche frontale untere Incisur ist der hyperplastische Muschelkopf nach vorne deutlich abgesetzt.

Abb. 8 Wie Abbildung 6, sagittale Incisur die bis knapp vor das hintere Ende der Concha media in 2 Teilmuscheln trennt.

Abb. 9 Wie Abbildung 6, am Ende des Muschelkopfes strahlt eine frontal verlaufende Furchen in die sagittale ein und setzt so den flächigen Muschelkopf deutlich von der Muschel ab.

Abb. 10 Rechte Seite von Abbildung 6, vergrößerter Kopf von Eihmurturbinale I mit Grübchen an der medialen Fläche.

Abb. 11 Rechte Nase: Incisur an der lateralen Fläche eines Eihmurturbinales I am Ende des Muschelkopfes.

Abb. 12 Linke Nase: Eihmurturbinale II mit zarter Furchenbildung am freien Rand, wahrscheinlich als Folge eines chronischen Hirndrucks.



1



2



3



4



5



6



7



8



9



10



11



12

ethmoidal avec formations de sillons au bord libre de l'ethmoïde. Jusqu'à la tuméfaction, voire la pulsation de la muqueuse à l'ethmoïde supérieur accompagné de pression cérébrale chronique

SUMMARY

The ethmoturbinals can be examined with precision by nasal endoscopy. The numerous anatomical variations deriving from the ethmoidal development are therefore of clinical importance: the cell in the concha a bead, the concha sinus, the rim or incision of the first ethmoid concha, the number of conchae, the partial development and the concha rudiments. Pathological changes can be identified only with profound knowledge of the anatomical variations. They include the waist at the free end of the concha as a sick cell of a concha, the mucosal prolapse out of a concha sinus with rim formation at the free end of the concha, mucous swelling or pulsation in the superior concha with chronic intracranial pressure.

LITERATUR

- André J. M. 1905 *Contribution à l'étude des lymphatiques du nez et des fosses nasales*. Thèse de Paris.
- Gegenbaur C. 1880. Ein Fall von mangelhafter Ausbildung der Nasenmuscheln. *Morph. Jb* (Leipzig) 5 191.
- Gruswald, L. 1917 Die Nasenmuscheln des Menschen. *Amer. Heft* (Wiesbaden) I 164 359.
- 1925 Deskriptive und topographische Anatomie der Nase und ihrer Nebenhöhlen. Denker Kahler in *Handbuch der Hals-Nasen-Ohrenheilkunde*. Bd. 1 J. Springer Berlin u. J. F. Bergmann, München.
- 1920. Der Seitenraum der Nase, dargestellt auf Grund der Entwicklung und des Vergleichs. *Arch. Laryng. Rhinol* (Berlin) 33 361.
- Hochstetler F. 1944 Über die Art und Weise in welcher sich bei Säugetieren und beim Menschen aus der sogenannten Riechgrube die Nasenhöhle entwickelt. *Z. Anat. Entwicklungsgesch* 113 105.
- Hoffmann, E. & Thiel, W. 1956. Untersuchung von

meintlicher und wirklicher Abflüsse aus dem Subdural und Subarachnoidalraum. *Z. Anat. Entwicklungsgesch* 119 283.

Killian, G. 1895 Zur Anatomie der Nase nach embryonalen. *Arch. Laryng. Rhinol* (Berlin) 2 234.

— 1895 3 17.

— 1896. 4 1.

Messerklinger W. 1970. (1) Die Endoskopie der Nase. *Mach. Ohrenheilk* 104 451.

— (2) Technik und Möglichkeiten der Nasendoskopie. *HNO* 20 133.

— (3) Nasendoskopie: Der mittlere Nasengang und seine unipertischen Entzündungen. *HNO* 20 112.

Peter K. 1913 *Atlas der Entwicklung der Nase und des Gaumens beim Menschen mit Ebruch der Entwicklungsstörungen*. G. Fischer, Jena.

— 1925 Vergleichende Anatomie und Entwicklungsgeschichte der Nase und ihrer Nebenhöhlen. Denker Kahler in *Handbuch der Hals-Nasen-Ohrenheilkunde*. Bd. 1 J. Springer Berlin u. J. F. Bergmann, München.

Richter H. 1932. Die normale Entwicklung der menschlichen Nase, in Sonderheit der Siebbeinzellen. *Arch. Ohr Nas Kehlkopfheilk* 131 265.

Spuler A. zit. n. H. Richter.

Starck, E. 1965 *Embryologie*. G. Thieme, Stuttgart 2. Aufl.

Zockerkandl, E. 1893 Normale und pathologische Anatomie der Nasenhöhle und ihrer paranasalen Anhangs. Ab. 1 W. Braumüller Wien-Leipzig 2. Aufl.

Zwilling H. 1912. Die Lymphbahnen des oberen Nasenabschnittes und deren Beziehungen zu den perimeningealen Lymphknoten. *Arch. Laryng. Rhinol* (Berlin) 26 66.

— 1914 Experimentelle Untersuchungen zur Mechanik der Intrakraniellen und zerebralen Komplikationen der Stirnhöhlenentzündungen. *Arch. Laryng. Rhinol* (Berlin) 28 271.

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EFFECT OF TESTING TECHNIQUE ON NASAL PRESSURE VARIATION

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Abstract The effect on nasal pressure change of breathing through a mask was studied in supine position by anterior rhinomanometry. Irrespective of the effect of shrinking drugs or the anatomical or functional health of the nose a significant increase was observed in both inspiratory pressure fall and expiratory pressure rise when testees were breathing through a mask. In a majority of cases, an increase in pressure change was observed leading to a different clinical result. Intensive studies of all technical details is recommended before a testing technique is adapted.

Of the available tests of nasal respiratory function, rhinomanometry has gained increasing clinical attention and acceptance during recent years. The techniques applied for this purpose are numerous and several parameters can be measured. The effect on pressure of the nose, a fall during inspiration and a rise during expiration, is easiest to record and accordingly is the parameter most often determined. On the other hand, a great variability in results of pressure rhinomanometry due for instance to the nasal cycle, has aroused considerable criticism against this test method (Williams et al., 1970).

Even the effect of introducing the testing device into the nose has been claimed to be significant and a source of error (Runderantz, 1970). In the present study the effect of placing a mask on the face of the testee has been analysed, as, according to our preliminary observations, this could be a source of variability in certain individual tests.

In this paper only the effects on nasal pressure of breathing through the mask are considered. An anterior rhinomanometry tech-

nique was used, due to the fact that in all attempts with a posterior recording the accumulation of secretion into the mouth made the test unreliable, because of several interruptions during the relatively long testing time.

The following technique was applied. A catheter, a standard No. 10 oxygen catheter with a sponge tip, was fixed airtightly to the contralateral nostril. The tip of the catheter was made airtight with a mixture of paraffin oil and vaseline, and fixation was completed with tapes. In this way an optimally stable fixation of the pressure detector was gained. This is proved by the few statistically significant changes recorded between the first and last testing period. The catheter was connected to an electromanometer calibrated for rhinomanometry. The testee remained supine during the whole test period and was asked to breathe quietly.

Recordings were made in several periods. In the first phase, a primary recording was taken when the testee appeared to have got used to bearing the catheter. Each recording period lasted for no less than 5 consecutive breaths, usually from 8 to 12. Half a minute later a secondary period was recorded and during continuous recording a mask was placed on the testee's face. This began the second phase with an immediate recording period of breathing through the mask. The mask was a simple diver's mask designed for a flow test so that it did not distort the external nose

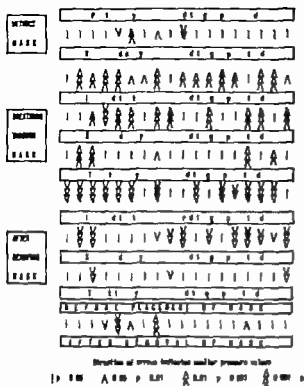


Fig. 1. Mean inspiratory pressure changes between consecutive recording periods and in the beginning and at the end of the test. Vertical lines represent one continuous test.

and yet fitted airtightly on the face (Kortekangas, 1971). Two additional periods were recorded while breathing through the mask.

A secondary recording period after a half minute pause and the tertiary recording period after a 1 minute pause. Recording continued when the mask was lifted from the face and this moment initiated the third phase with an immediate recording period after removing the mask. Again two control recording periods were taken: the secondary recording period after a half minute pause and the tertiary recording period after a 1 minute pause.

The mean respiratory pressure falls in relation to ambient pressure of each breath of the above periods were statistically compared applying Student's *t*-test. The results from 18 testees, subjects with pathological and normal noses, are given together due to the fact that no differences in reaction could be traced as related to the nasal state.

RESULTS AND DISCUSSION

In Fig. 1 mean inspiratory pressure changes in this trial are given. There are significant changes in the mean inspiratory pressure fall in four cases between the primary and secondary period without mask at the beginning of the test with the testee breathing through one nostril, an increase in two cases and a decrease in two. Considering the frequency of variation in nasal pressure changes this is an unexpectedly low incidence and is certainly due to the fact that most of these tests were made at a time when the effect of an earlier shrinkage of the mucosa still persisted.

When mean inspiratory pressure changes in the beginning and at the end of the whole test were compared, a significant change was seen in five cases, a pressure fall in two and a pressure rise in three. This result constitutes the main proof of the validity of this test, which revealed a pressure rise when the testee was breathing through a mask and pressure fall when the mask was withdrawn. This change could appear immediately during the first breath or a few moments later. The moment of appearance of this reaction seems to be individual, but very often either the immediate or the delayed type was characteristic both under the mask in the rise and after its withdrawal in the fall.

As the results of consecutive expiratory changes between all the periods of the test were very similar to those of the inspiratory test, the expiratory results are not given in detail.

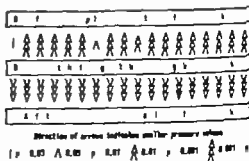


Fig. 2. Mean inspiratory pressure changes.

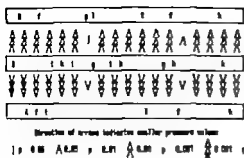


Fig. 3. Mean expiratory pressure changes.

Figure 1 records only the significance of changes between adjacent recording periods and a slow reaction may thus be hidden. For this reason, a comparison of mean pressure fall over three phases, before placement of the mask, during breathing through the mask and after its withdrawal, are given in Fig. 2. The corresponding pressure rises during expiration are given in Fig. 3. This result shows that breathing through a mask in supine position causes a significant increase in both inspiratory and expiratory nasal pressure changes.

When the change is considered in terms of normal nasal respiratory pressure change (< 15 mm/H₂O), elevated nasal pressure change (16–30 mm/H₂O) or highly elevated pressure change (> 31 mm/H₂O) the following result was obtained. In 8 testees, the pressure remained normal during the whole test. In 10 testees, nasal pressure change was normal without the mask but became elevated when the testee was breathing through the mask. In one testee, the mean pressure rise reached 58 mm/H₂O. In one testee, pressure remained elevated during the whole test.

This observation cannot be ignored in the evaluation of rhinomanometric pressure tests. Before any specific testing technique can be

recommended for general use, intensive studies should be made on each detail of the procedure involved.

RÉSUMÉ

On a utilisé la technique du masque dans la rhinomanométrie antérieure. Les changements dans les variations de la pression nasale sont analysés du point de vue de la respiration libre et la respiration par un masque.

ZUSAMMENFASSUNG

Die Wirkung der Maskenatmung am rhinomanometrischen Drucktest bei liegenden Versuchspersonen ist untersucht worden. Eine anteriore Rhinomanometrie mit fixiertem Druckkatheter erwies sich stabil genug für wechselseitige Registrierung ohne und durch die Maske. Ein statistisch signifikantes Steigen sowohl der inspiratorischen als der expiratorischen Druckveränderungen konnte man unter Maskenatmung sehen, ohne dass Personen mit pathologischen Nasenveränderungen ein verschiedenes Verhalten von Normalpersonen aufwiesen. In der Mehrzahl der Fälle war diese Veränderung bedeutend genug, sodass die Resultate der Rhinomanometrie falsch als pathologisch durch die Maske und normal ohne sie zu beobachten war. Ein sorgfältiges Studieren aller Einzelheiten und deren Wirkung an den Resultaten ist notwendig bevor eine rhinomanometrische Methode empfohlen werden kann.

REFERENCES

- Kortekangas, A. E. 1971. Clinical application of rhinomanometry. *Rhinology* 9, 144.
- Rundcrantz, H. 1970. Discussion of paper Clinical Application of Rhinomanometry of A. E. Kortekangas. *Proc. IV European Congr. Rhinology* Odense, Denmark.
- Williams, H. L. et al. 1970. *Definition of Terms Used in Rhinomanometry with Suggested Standard Symbols*. American Academy of Ophthalmology and Otolaryngology, Rochester, Minnesota 55901 USA.

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ON THE ORIGIN OF POSITIONAL ALCOHOL NYSTAGMUS

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Abstract. The vestibular system plays an important role in the origin of positional alcohol nystagmus (P.A.N.). An investigation was performed on the effect of artificial lesions in the peripheral vestibular organs of rabbits upon P.A.N. In rabbits in which the otooliths were eliminated on both sides, a P.A.N. could not be provoked. If on one side the labyrinth was totally destroyed, a P.A.N. could still be elicited, but if in the only intact labyrinth the utricular nerve was also transected a P.A.N. could no longer be elicited. That the otooliths are responsible for the P.A.N. also finds support in the fact that increased g forces effect a P.A.N. in rabbits as well as in man.

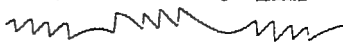
Positional alcohol nystagmus following administration of alcohol to animals was demonstrated for the first time in 1842 (Flourens). Bárány (1911) and Bárány & Rothfeld (1913) were the first to perform experimental studies in order to evaluate the effect of alcohol upon the vestibular system.

Since positional alcohol nystagmus has been described in experimental studies in animals and man by various authors (Goldberg, 1941 1943 1966 Plenkera, 1943) In 1956 nystagmographic studies were performed for the first time (Aschan et al., 1956, 1965 Aschan & Bergstedt 1957) Positional alcohol nystagmus appears in two phases (Aschan, 1958 Walter 1954) The first phase (PAN I) appears 20-30 minutes after the intake of alcohol and shows a nystagmus with the fast component beating in the direction of the side position i.e. in left-side position to the left and in right side position to the right. The first phase lasts about 3 hours. 1 $\frac{1}{2}$ -2 hours later the second phase (PAN II) appears when the

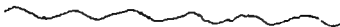
subject is in lateral position but the direction of this nystagmus is opposite to the direction of PAN I. PAN II goes on up to 10 hours after the intake of alcohol. PAN can only be elicited if there is an intact vestibular apparatus, after bilateral labyrinthine destruction PAN can no longer be provoked, neither in rabbits (De Kleyn & Versteegh, 1930) nor in humans (Fregly & Graybiel, 1968 Harris et al. 1962) De Kleyn & Versteegh (1930) described that extirpation of the sacculle did not have any effect on the appearance of PAN (1930) after labyrinthectomy on one side, alcohol nystagmus could still be provoked but in the side position opposite to the operated ear only Nito et al. (1964) found in experiments in cats that PAN does not occur following the inactivation of all six semicircular canals. This indicates that the semicircular canals are necessary to elicit PAN. Mooney et al. (1965) inactivated both the horizontal semicircular canals in cats, after which a horizontal alcohol nystagmus could no longer be elicited in any position, but the vertical and rotatory components of the nystagmus were still present. These authors state that positional alcohol nystagmus is initiated by the action of gravity on receptors of the semicircular canals. However the functional control tests in their experiments were not very accurate.

Hill et al. (1972) found no consistent effects of alcohol on either the vertical nystagmus or the vertigo produced by coriolis stimulation.

A. TORSION-SWING TEST



LEFT SIDE POSITION



B. PARALLEL SWING TEST

RIGHT SIDE POSITION



LEFT SIDE POSITION



C. P.A.N.

RIGHT SIDE POSITION



Fig 1 Normal rabbit. A No preponderance. B Equal sinusoidal eye-movements in both side positions. C Equal P.A.N. in both side positions.

Decrease in turning sensations and in nystagmic responses to angular acceleration were found and most evident during the first hour or two after drinking. This last finding points to an effect of alcohol on the processing of information from the semicircular canals. The review of the literature shows that the investigators do not agree on the origin of positional alcohol nystagmus. In order to gain more insight on the origin, we investigated the alcohol nystagmus in rabbits which had artificial lesions of the peripheral vestibular apparatus.

These rabbits were normal laboratory animals, all in good health, free from spontaneous or positional nystagmus. They had no directional preponderance in the torsion-swing test, equal reactions in the parallel-swing test, this means sinusoidal eye movements in left and right-side position when swinging sideways, as well as in the direction of the longitudinal body axis, and an equal irritability of the labyrinths in the caloric test.

Six groups of 10 rabbits each were subjected three times to experiments in which alcohol nystagmus was provoked by means of intra-

peritoneal administration of ethyl alcohol. The eye movements in the horizontal plane were recorded nystagmographically. The recordings with the animals were made in total darkness. The rabbits collected in one group all had a specific lesion of the vestibular apparatus.

Group I consisted of normal rabbits. Group II consisted of rabbits which were labyrinthectomized on one side in group III the rabbits were labyrinthectomized on both sides. The utricular nerve was transected on one side in the rabbits of group IV and both the utricular nerves were transected in the rabbits of group V. In the last group VI rabbits were labyrinthectomized on one side and utriclectomized on the other side.

Labyrinthectomies were performed in the way described by Versteegh (1927). The surgery on the utricular nerve was done by the same approach. Surgery of two ears was always performed in two steps, one step for each ear with an interval of 4 weeks. The results of the surgery were functionally controlled, the labyrinthectomies by the non-irritability to caloric testing and a directional

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subject is in lateral position but the direction of this nystagmus is opposite to the direction of PAN I. PAN II goes on up to 10 hours after the intake of alcohol. PAN can only be elicited if there is an intact vestibular apparatus, after bilateral labyrinthine destruction PAN can no longer be provoked, neither in rabbits (De Kleyn & Versteegh, 1930) nor in humans (Fregly & Graybiel, 1968; Harris et al., 1962). De Kleyn & Versteegh (1930) described that extirpation of the sacculi did not have any effect on the appearance of PAN (1930) after labyrinthectomy on one side, alcohol nystagmus could still be provoked but in the side position opposite to the operated ear only. Nito et al. (1964) found in experiments in cats that PAN does not occur following the inactivation of all six semicircular canals. This indicates that the semicircular canals are necessary to elicit PAN. Money et al. (1965) inactivated both the horizontal semicircular canals in cats, after which a horizontal alcohol nystagmus could no longer be elicited in any position, but the vertical and rotatory components of the nystagmus were still present. These authors state that positional alcohol nystagmus is initiated by the action of gravity on receptors of the semicircular canals. However the functional control tests in their experiments were not very accurate.

Hill et al. (1972) found no consistent effects of alcohol on either the vertical nystagmus or the vertigo produced by coriolis stimulation.

A. TORSION SWING TEST



LEFT SIDE POSITION

B. PARALLEL SWING TEST

RIGHT SIDE POSITION

LEFT SIDE POSITION

C. V.A.B.

RIGHT SIDE POSITION

Fig. 4 Rabbit in which both the utricular nerves are cut through. A No directional preponderance. B No response. C No response.

ml ethanol 96% per kg bodyweight was administered intravenously in a 20% diluted solution with normal saline. In most cases PAN was noticed to appear within 10 minutes after the administration of alcohol. In some experiments higher g-values were applied. This was performed in the way described in earlier publications (Oosterveld & van der Laarse, 1969; Oosterveld, 1970).

RESULTS

Group I

The normal rabbits showed a PAN in both side positions. In 6 out of 10 rabbits there was a small difference in frequency between the nystagmus to the right and to the left, but always less than 10% (Fig. 1).

Group II

In the rabbits with the unilaterally destroyed labyrinth the alcohol nystagmus was present when the rabbits were in the side position on the intact labyrinth. The other side position provoked in 6 rabbits a very slight nystagmus but in the remaining rabbits no nystagmus at all (Fig. 2).

Group III

None of the rabbits with the bilaterally destroyed labyrinths presented a PAN after the application of alcohol. Even with doubled dosage no PAN could be provoked, nor when the rabbits were subjected to higher g-values, up to 3-g.

Group IV

In the rabbits with the unilateral utriclectomy a clear PAN could be provoked when the animals were put in the side position opposite to the operated ear. Only 2 rabbits out of 10 showed some nystagmus beats when put in the side position on the operated side (Fig. 3).

Group V

None of the bilaterally utriclectomized rabbits of group V showed any nystagmus after the alcohol administration. No vertical or rotatory movements were noted. The rabbits of this group were subjected to higher g-values, up to 3-g. but this condition had no effect on the findings. Even an increase of alcohol dose with a factor 2 up to 4 cc/kg did not provoke a nystagmus (Fig. 4).

A TORSION SWING TEST

LEFT SIDE POSITION

B PARALLEL SWING TEST

RIGHT SIDE POSITION

LEFT SIDE POSITION

C P.A.N.

RIGHT SIDE POSITION

Fig. 5 Rabbit in which the labyrinth on the right side is destroyed and the utricular nerve on the left side is transected. *A* Nystagmus preponderance for

a nystagmus to the left. *B* No response. *C* No response.

Group VI

The rabbits of the last group had one labyrinthectomized ear and one sectioned utricular nerve on the other side. None of these animals showed a PAN nor when the alcohol

dosage had been doubled. Here neither vertical nor rotatory eye-movements were noted (Fig. 5) Table I shows the combined results of all the experiments.

Table I

	RESPONSE ON VESTIBULAR TESTS						P.N.	ANNO
	ROTATION TEST		LINEAR ACCELERATION TEST		CALORIC TEST		P.N.	ANNO
	RESPONSE		RESPONSE		RESPONSE			
	NO STIMULATION PERMANENT IMPACT STIMULATED PERMANENT IMPACT	NO STIMULATION PERMANENT IMPACT STIMULATED PERMANENT IMPACT	NO STIMULATION PERMANENT IMPACT STIMULATED PERMANENT IMPACT	NO STIMULATION PERMANENT IMPACT STIMULATED PERMANENT IMPACT	WET DRY ONE EAR	WET DRY ONE EAR		
STIMULATED	•		•		•		•	
(APPROXIMATELY ON ONE SIDE)		•		•		•		
APPROXIMATELY ON BOTH SIDES			•		•		•	
STIMULATED ON ONE SIDE	•			•		•		•
STIMULATED ON BOTH SIDES	•				•	•		•
STIMULATED ON ONE SIDE AND LABYRINTHECTOMIZED ON THE OTHER SIDE		•			•	•		

CONCLUSION

Rothfeld (1913) pointed to the regularity of the phenomenon of positional alcohol nystagmus observed in lateral positions of the head.

The results of our experiments point to the conclusion that the utricle is an essential organ for the genesis of positional alcohol nystagmus.

In a previously published paper (Oosterveld, 1970) we described the fact that higher g values, which means an otolithic stimulation, were able to arouse a PAN even if the amount of administered alcohol was very small and even if the moment of the application of alcohol was more than 12 hours prior. In weightlessness, a PAN disappears immediately. These findings also point in the direction of a specific role of the otoliths in the genesis of alcohol nystagmus. Nevertheless, a nystagmus is not the usual response following otolithic stimulation.

In a previous publication the hypothesis was stated that the otoliths modulate a centrally provoked nystagmus both in amplitude and direction depending on the position in space. During intoxication with alcohol a nystagmus tendency is continuously present and it is only released in certain positions. These views agree with those of Aschan et al. (1964, 1965) and Grant et al. (1964) that the place of the origin of PAN must be searched more centrally.

The otoliths or the utricle seem to play an essential role in the trigger mechanism of the appearance of alcohol nystagmus.

RESUME

Le nystagmus vestibulaire joue un rôle très important dans l'origine du nystagmus de position provoqué par intoxication alcoolique (P.A.N.). Une investigation a été faite chez le lapin concernant l'effet des lésions artificielles labyrinthiques sur le P.A.N. Avec seulement les canaux semi-circulaires intacts il était possible de produire un P.A.N. Chez les lapins ayant subi une destruction labyrinthique unilatérale on pouvait encore provoquer le P.A.N. Si par contre on sectionnait le nerf utriculaire du côté non opéré il était de nouveau impossible de produire le P.A.N. Le fait que la partie statique du système

vestibulaire est responsable pour le P.A.N. est confirmé par l'expérience que le P.A.N. est fortement influencé par des accélérations linéaires aussi bien chez le lapin que chez l'homme.

ZUSAMMENFASSUNG

Der Vestibularapparat spielt eine grosse Rolle beim Entstehen eines Alkohol-Lage-Nystagmus (P.A.N.). Eine Untersuchung wurde gemacht zur Beurteilung des Effekts künstlich hergestellter Läsionen des peripheren Vestibular Organs bei Kaninchen. Wenn nur die Bogengänge noch intakt waren, war es nicht mehr möglich ein P.A.N. zu erzeugen. Bei Kaninchen mit einseitiger Labyrinth-Zerstörung war es aber möglich einen P.A.N. zu erzeugen. Wenn aber bei diesen Tieren der Nervus utricularis an der Seite der Labyrinth-Zerstörung durchschnitten worden war war es nicht mehr möglich einen P.A.N. zu erzeugen. Dass der statische Teil des Gleichgewichtsorgans verantwortlich ist für den Alkohol-Lage-Nystagmus wird auch bestätigt durch den Befund, dass geringfügige Beschleunigungen in hohem Masse einen P.A.N. beeinflussen können, bei Kaninchen so wie auch bei Menschen.

REFERENCES

- Aschan, O. 1958. Different types of alcohol nystagmus. *Acta Otolaryng* (Stockh.), Suppl. 140 69.
— 1967. Habituation to repeated rotatory stimuli (cupulometry) and the effect of antineurotic drugs and alcohol on the results. *Acta Otolaryng* (Stockh.) 64 95.
Aschan, O. & Bergstedt, M. 1957. Balancinkönsfunktion vid alkoholintoxikation. Nystagmografiiska undersökningar. *Ann Acad Regiae Sci Upsalien* 1 36.
Aschan, O., Bergstedt, M. & Goldberg, L. 1964. Positional alcohol nystagmus in patients with unilateral and bilateral labyrinthine destruction. *Confin Neurol* 24 80.
Aschan, O., Bergstedt, M., Goldberg, L. & Lennell, L. 1965. Positional nystagmus in man during and after alcohol intoxication. *Quart J Stud Alcohol* 17 381.
Aschan, O., Bergstedt, M. & Stahle, J. 1956. Nystagmography. *Acta Otolaryng* (Stockh.), Suppl. 129.
Aschan, O., Ekvall, L. & Grant, G. 1964. Nystagmus following stimulation in the central vestibular pathways using permanently implanted electrodes. *Acta Otolaryng* (Stockh.), Suppl. 192 11.
Bárány, R. 1911. Experimentelle Alkoholfunktion. *Machr Ohrenheilk* 45 959.
Bárány, R. & Rothfeld, J. 1913. Untersuchungen des Vestibularapparates bei akuter Alkoholfunktion und Delirium tremens. *Z Nervenhk* 50 133.
Bergstedt, M. 1961. Studies of positional nystagmus in the human centrifuge. *Acta Otolaryng* (Stockh.), Suppl. 165 1.
Flourens, P. 1842. *Recherches expérimentales sur les propriétés et les fonctions du système nerveux*. (2nd ed.) Paris.

A TORSION SWING TEST

LEFT SIDE POSITION

B PARALLEL SWING TEST

RIGHT SIDE POSITION

LEFT SIDE POSITION

C P.A.N.

RIGHT SIDE POSITION

Fig 5 Rabbit in which the labyrinth on the right side is destroyed and the utricular nerve on the left side is transected. A Nystagmus preponderance for

a nystagmus to the left. B No response. C No response.

Group 11

The rabbits of the last group had one labyrinthectomized ear and one sectioned utricular nerve on the other side. None of these animals showed a PAN nor when the alcohol

dosage had been doubled. Here neither vertical nor rotatory eye-movements were noted (Fig. 5). Table I shows the combined results of all the experiments.

Table I

E X P E R I M E N T	RESPONSE ON VESTIBULAR TESTS										P.A.	
	ROTAT ON TEST			LINEAR ACCELERATION TEST			CALORIC TEST			PROMPT	ASSYST	
	RESPONSE		NO RESPONSE	RESPONSE		NO RESPONSE	RESPONSE		NO RESPONSE			
	NO NYSTAGMUS FOR BALANCE	STEEPLE PREPONDANCE		NO PREPONDANCE EFFECT OF 100 PM (100)	PREPONDANCE EFFECT OF 100 PM (100)		NO NYSTAGMUS	NO NYSTAGMUS		NO EFFECT OF 100 PM (100)	NO EFFECT OF 100 PM (100)	
NORMAL	•			•			•			•		
Labyrinthectomized on one side		•			•			•			•	
Labyrinthectomized on both sides			•			•			•			•
Utriclectomized on one side	•				•		•				•	
Utriclectomized on both sides	•				•		•					•
Utriclectomized on one side and labyrinthectomized on the other side		•			•		•					•

EFFECT OF REPETITIVE OPTOKINETIC STIMULATION UPON
OPTOKINETIC AND VESTIBULAR RESPONSES

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Abstract. The question of whether habituation of optokinetic nystagmus occurs with repetitive stimulation was studied in a series of trials in 15 human subjects. The results indicate that there is no response decline characteristic of habituation, but on the contrary a constant and definite increase of optokinetic response, interpreted as the result of a positive learning process. The latter does not influence or modify the caloric, rotatory and galvanic vestibular responses.

Repetitive stimulation of a sensory organ may induce *habituation* which, according to the definition of Thorpe (1950) represents a simple process of learning not to respond to stimuli, which tend to be without significance in the life of the organism. The term of habituation, introduced by Abels (1906), is generally used to indicate the phenomenon of a progressive response decline to repeated stimuli, having identical parameters (Monnier et al., 1970). Habituation of the vestibular system by various sorts of stimuli (rotatory caloric) has been reported by many authors (Hood & Pfaltz, 1954; Henriksson et al., 1961; Fluor & Mendel, 1969; Guedry, 1964; Forssman, 1964; Forssman et al., 1963; Brown & Crampton, 1965; Collins, 1965; 1966; Aschan, 1967; Pfaltz & Piffko, 1970; 1970; Monnier et al., 1970; Montandon et al., 1970; Greiner et al., 1970; Pialoux et al., 1970; Kaphan, 1970). However reports on the habituation of optokinetic nystagmus are scarce and contradictory (Fernández & McClure (1963)

could not confirm the presence of a progressive decline of optokinetic responses in man following repetitive optic stimulation. On the other hand, Oku (1954), Motohashi (1965), Shtrakt (1967) and Hinoki et al. (1971), based upon animal experiments, reported a gradual increase of optokinetic responses after repeated optic training.

The question arises, whether any modification of the optokinetic responses induced by repeated optic stimulations of varying intensity and direction, may be observed in normal human individuals. If such a modification of the optokinetic responses could be confirmed, it would involve the question whether this modified response were the result of *habituation* or of some other learning-mechanism. Another question is, whether vestibular responses might be modified by repeated optokinetic stimulation. We have made an attempt to approach this very complex problem by carrying out the following experimental study.

Test procedure

The observations were made on 15 healthy well motivated subjects, with an age range from 14 to 45 years. None of the subjects had any gross abnormality of vision or eye motility.

The instrumentation used for eliciting optokinetic nystagmus consisted of a Bárány type drum (B.T.D.) on the one side (Fig. 1) and a Jung-type projector-screen (J.T.P.S.) on the other side (Fig. 2). The former was used

This work was supported by the Schweizerischer Nationalfond zur Förderung der wissenschaftlichen Forschung.

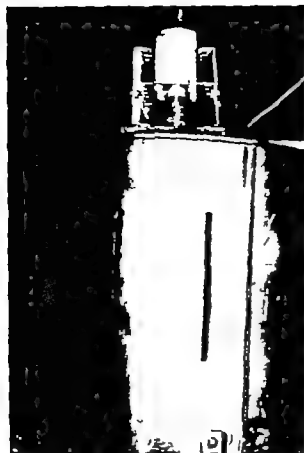


Fig. 1. Bárány Type Drum. This apparatus is a modification of the optokinetic stimulator designed by J. Lang (1962), consisting of a metal box, which contains a metal cylinder driven by an electromotor. On the case of the cylinder 10 circular perforations (2 cm diameter) are set at equal distance, illuminated by an electric lamp from the inside. They produce well-circular light spots on a ground glass screen, on a rectangular window (2.5–10 cm), which is on the front side of the metal box. The

distance between the circular perforations implies that only one light spot will appear on the screen at any time. The test subject, whose head is fixed on a metal support (distance head stimulator 57 cm) is asked to follow the light spots passing in front of him. With this stimulating device only foveolar optokinetic nystagmus is induced because merely one target (light spot) is moving across the test subject's visual field at any time.

for eliciting foveolar nystagmus by narrow angle-target stimulations, the latter for eliciting both foveolar and peripheral (retinal) optokinetic nystagmus (OKN) by wide-angle target stimulation. Foveolar OKN is induced by an angular velocity of the wide-angle tar-

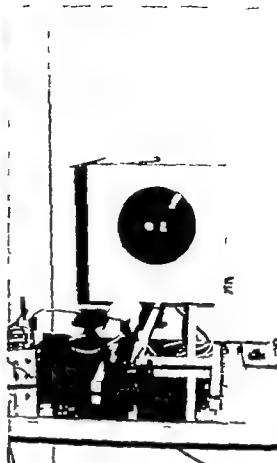


Fig. 2. Jung Type Projector Screen. A metal cylinder with 12 slots, set at equal distance (30°) drives an electromotor in projecting dark and light strips on a white half cylindrical screen (diameter 160 cm). The projector is located and rotated above the head of the test subject, who is in front of the screen a distance of 80 cm. The length of the projected stripes is 80 cm and the width 3 cm, corresponding to a visual angle of 57° and 2.1° respectively.

gets <90°/sec, whereas any target speed >90°/sec will initiate only peripheral OKN (Miyoshi, 1970).

Recording of eye movements: different photoelectric nystagmography (Pfalz & Richter 1955–1956) and by means of a special d.c. recording design (Richter et al., 196) which is free of deformities of the tracings.

Habituation test series

One trial, repeated on 10 successive days, consisting of 10 successive stimulations daily.

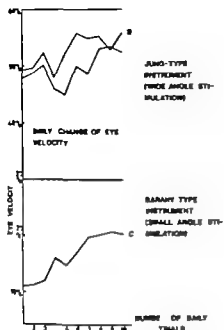


Fig. 3 (a) Daily change of eye velocity (velocity of the slow phase of optokinetic nystagmus). Groups A and B (wide angle stimulation) show an irregular but definite daily change of eye velocity resulting in a final increase of optokinetic responses, comparing the results before and after the habituation trials. Group C (small angle habituation) shows a regular definite and constant daily change of eye velocity also resulting in a final increase of optokinetic responses.

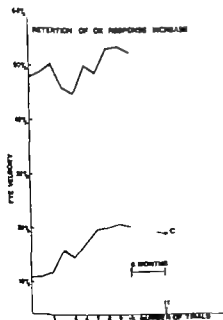


Fig. 3. (b, c) Retention of OK response increase. In Group B there is no conclusive evidence of retention, whereas in Group C optokinetic responses 6 months

15 sec duration, each followed by a 15 sec recess.

Target movement velocity: 30 /sec for B.T.D.-stimulation corresponding to a small angle stimulation, and 60 /sec for J.T.P.S. stimulation, corresponding to a wide angle stimulation.

Before and after each trial the following tests were carried out: caloric test (Hallpike-Fitzgerald method)—stepping test—optokinetic test with target velocities of 15 /sec, 20 /sec, 25 /sec, 30 /sec for B.T.D. and J.T.P.S. stimulation and 60 /sec, 90 /sec, 120 /sec for J.T.P.S.-stimulation (bidirectional stimulation).

As a parameter for the evaluation of the optokinetic response we have used the velocity of the slow phase.

The 15 test persons were divided into 3 groups (A, B, C) of 5 individuals each.

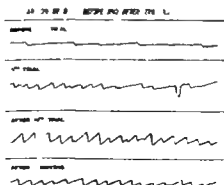
Group A: Habituation by J.T.P.S.-stimulation towards the right (direction of target movement).

Group B: Habituation by J.T.P.S.-stimulation towards the left.

Group C: Habituation by B.T.D.-stimulation towards the right.

DISCUSSION OF RESULTS

The curves in Fig. 3 a and b reproduce the average slow phase velocity values of 5 per



after the last trial are still exhibiting a marked increase (3).

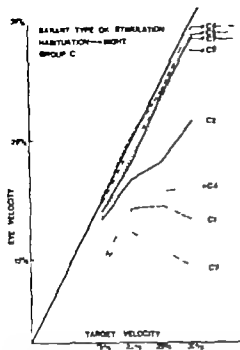


Fig. 4 Habituation to the right by small angle optokinetic stimulation (target velocity 30°/sec.). The target velocity for the control tests is 15/sec-20°/sec 25 sec-30°/sec.

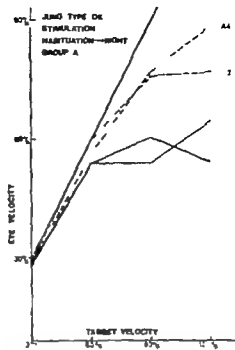


Fig. 5 Repetitive wide angle optokinetic stimulation by a Jung-Type Projector Screen Equipment" (JTPS). For the habituating trials a target velocity of 60°/sec is used, for the control tests a target velocity of 30°/sec, 60°/sec, 90°/sec, and 120°/sec.

Figs. 4 and 5.

A/B-1 C-5 control test before trial 1	} Optokinetic stimulation → R by JTPS
A/B-2/C-6: control test after trial 10	
A/B-3/C-7 control test before trial 1	} optokinetic stimulation → L by JTPS
A/B-4 C-8 control test after trial 10	
A/B-5/C-1 control test before trial 1	} optokinetic stimulation → R by LTD
A/B-6 C-2: control test after trial 10	
A/B-7 C-3 control test before trial 1	} optokinetic stimulation → L by LTD
A/B-8 C-4 control test after trial 10	

sons belonging to the same group. Optokinetic responses following repeated optic stimulation do not exhibit a response decline but on the contrary a significant response increment. This may be seen from Fig. 3 a showing the daily change of the slow phase velocity of optokinetic nystagmus, and also from Fig. 3 b demonstrating a maintenance of this response increment for 6 months after the last trial.

It is interesting to recognize that the establishment of this response increment is independent of the direction and the varying velocity of the optic training movement. E.g. test subjects habituated to the right were

showing an increase of both left and right optokinetic responses, even if different target velocities were used during the trials and for the control tests. Both findings may be interpreted as the result of a transfer-mechanism.

Caloric test results did not reveal any conclusive changes after repetitive optic stimulation although the total reaction seems to show some tendency to increase. However further investigations are necessary to come to a final conclusion.

The galvanic test results before and after 10 OAN-trials do not demonstrate any definite response decay or increment, nor the establishment of a directional preponderance,

which might be related to the direction of the optic training movement.

Stepping test

Subjects show an evident tendency to rotate in a direction opposite to the direction of the optic stimulation, applied during the daily trial and corresponding to the quick phase of OKN. In subjects belonging to the groups tested by wide-angle stimulation (Groups A and B) this observation was even more evident and more constant than in Group C, stimulated by small-angle stimulation.

In our experimental study on 15 normal subjects, repeated unidirectional optokinetic stimulations did not result in an *acquisition* of a progressive optokinetic response decline a finding which is in agreement with the observations made by Fernández & McClure (1963). On the contrary we observed the opposite phenomenon, i.e. the *acquisition* and *retention* of a very definite response increment (Fig. 3 a, b). Similar observations have already been made by Oku (1954), Hinoki & Terayama (1966) and by Shuraki (1967) in animal experiments. The test procedure of Fernández & McClure (1963), in which they did not observe any response increase, was quite different from ours (constant angular velocity of optokinetic stimulus instead of different stimulation intensities—exposure to only one trial instead of exposure to several trials over a certain period of time). For that reason our results cannot be compared with theirs.

The question arises, whether the increment of the optokinetic response may be interpreted as a true *habituation phenomenon*.

Habituation is a decrement of response with repeated stimulation. It differs from both sensory adaptation and nerve accommodation in its characteristics, and does not depend on the effector fatigue (Sharpless & Jasper 1956; Hernández-Peón & Brust-Carmona, 1961). It is interpreted as the simplest process of *negative learning*.

The occurrence of a *response increase* following repeated optic stimulations of varying

intensity cannot be interpreted by habituation. This is in agreement with a statement made by Kornhuber (1966) indicating that habituation of optokinetic nystagmus does not exist.

Optokinetic mechanisms are greatly influenced by alertness. When a small object moves through the visual field of a person, it induces a "grasp reaction" requiring attention (Monnier 1967). The underlying mechanism of this reaction is foveal vision and its resultant foveolar OKN. In our test series, OK response increment and its retention was more marked in subjects exposed to repetitive foveolar optic stimuli (group C, Fig. 4) than in subjects exposed to stimuli involving peripheral vision (groups A and B Fig. 5). According to Hood (1967) foveal OKN is under the controlling influence of the centre for voluntary gaze in the frontal cortex, while peripheral OKN has a more reflex character and comes under the influence of the occipital visual center. The modification of the OK response, more marked for its foveolar type, may therefore be due to a process of learning, occurring at the level of the cerebral cortex or some other central region of convergence of sensory impulses. If we are accepting Glaser's (1966) definition of learning (acquisition of new responses or a modification of preexistent responses, induced by storage of novel information) we may assume that the increase of optokinetic responses, induced by repetitive optokinetic stimulation or optic training, is the resultant of a positive *learning-process* probably serving the visual system to improve its ability to pick up the periodicity of simple motion patterns, in order to use this sensory information for an improvement of the motor output.

RÉSUMÉ

Le problème de l'habituation du nystagmus optocinétique et son influence sur les réponses vestibulaires a été étudié dans 15 sujets normaux, exposés à des stimulations optiques répétées. Le résultat de cette expérience démontre qu'il existe pas d'habituation optocinétique, caractérisée par une diminution des ré-

ponées nystagmiques dans le sens de d'un negative learning process" Par contre, une série de stimulations optiques répétées amène une augmentation nette des réponses optocinétiques, un phénomène qui est interprété comme résultant d'un positive learning process" Le dernier n'exerce aucune influence sur les réponses vestibulaires thermiques, rotationnelles et galvaniques.

ZUSAMMENFASSUNG

Das umstrittene Problem der Habituation oder Gewöhnung des optokinetischen Nystagmus wurde an 15 gesunden Individuen mittels wiederholter optischer Reizungen untersucht. Die Ergebnisse zeigten, dass es keine echte optokinetische Gewöhnung im Sinne der verminderten Reizantwort gibt, sondern dass im Gegenteil am Ende der Versuchsstreife regelmäßig eine erhebliche Steigerung der optokinetischen Reizantwort beobachtet werden konnte. Dieses Phänomen wurde als Ausdruck eines positiven Lernprozesses gedeutet, im Gegensatz zur Gewöhnung, welche den einfachsten Typ eines negativen Lernprozesses darstellt. Letzterer beeinflusst in keiner Weise die vestibulären Reizantworten.

REFERENCES

- Abels, H. 1906. Über Nachempfindung im Gebiete des kinästhetischen und statischen Sinnes. Ein Beitrag zur Lehre von Bewegungswandel (Drehschwindel). *Z Psychol Physiol Sinnesorg* 43 268.
- Achan, I. 1967. Habituation to repeated rotatory stimuli and the effect of antineurotic drugs and alcohol on the results. *Acta Otolaryng* (Stockh.) 64 95.
- Brown, J. H. & Crampton O. H. 1965. Vision and vestibular habituation. *Acta Otolaryng* (Stockh.) 61 80.
- Ilina, W. E. 1965. Subjective responses and nystagmus following repeated unilateral caloric stimulation. *Ann Otol* 74 1034.
- 1966. Problems in spatial orientation. Vestibular studies of figure skaters. *Trans Amer Acad Ophthalmol* 75 575.
- Fernández, C. & Risco-MacClure J. S. 1963. Studies on habituation of vestibular reflexes. V. The optokinetic nystagmus. *Ann Otol* 72 336.
- Fluur, E. & Mendel, L. 1969. Relation between strength of acceleration and duration of postacceleratory nystagmus. *Acta Otolaryng* (Stockh.) 68 127.
- Forrestman, B. 1964. Studies on habituation of vestibular reflexes. Habituation in light of calorically induced nystagmus laterotorsion and erlgo in man. *Acta Otolaryng* (Stockh.) 57 163.
- Forrestman, B., Henriksen, N. O. & Dolowitz, D. A. 1963. Studies on habituation of vestibular reflexes: Habituation in darkness of calorically induced nystagmus laterotorsion and erlgo in man. *Acta Otolaryng* (Stockh.) 56 663.
- Glaeser, E. M. 1966. Die physiologischen Grundlagen. *Acta Otolaryng* 75.
- der Gewöhnung (The physiological basis of habituation) Oxford University Press, London.
- Greiner, G. F., Courant, C., Maître, B., Collard, M. & Thiebaut, M. S. 1970. Etude de l'habituation vestibulaire par les stimulations pendulaires répétées. *Adv Oto-Rhino-Laryng* 17 136.
- Guedry, F. E. 1964. Visual control of habituation to complex vestibular stimulation in man. *Acta Otolaryng* (Stockh.) 58 337.
- Henriksson, N. O., Kobus, R. & Fernández, C. 1961. The effect of repetitive caloric tests. *Acta Otolaryng* (Stockh.) 53 333.
- Hernández Peon, R. & Brust-Carmona, H. 1961. Functional role of subcortical structures in habituation and conditioning. In *Brain mechanisms and learning* (ed. de la Fresnaye), C. C. Thomas, Springfield, Ill.
- Hinoki, M. & Terayama, K. 1966. Studies on the optokinetic nystagmus from the standpoint of body equilibrium. *Acta Otolaryng* (Stockh.) 62 8.
- Hinoki, M., Terayama, K. & Kashiwabara, S. 1971. Role of cervical proprioceptors for adaptation to optic stimulation. *Equilibrium Res Suppl.* 2 12.
- Hood, J. D. 1967. Observations upon the neurological mechanism of optokinetic nystagmus with special reference to the contribution of peripheral vision. *Acta Otolaryng* (Stockh.) 63 208.
- Hood, J. D. & Pfaltz, C. R. 1954. Observations upon the effects of repeated stimulation upon rotation and caloric nystagmus. *J Physiol* 124 130.
- Kaphan, G. 1970. A propos de l'habituation vestibulaire. *Adv Oto-Rhino-Laryng* 17 196.
- Kornhuber, H. H. & Spillmann, L. 1964. Zur visuellen Feldorganisation beim Menschen. *Pflügers Arch Physiol* 279.
- Laag, J. 1962. L'évocation du nystagmus optocinétique à l'aide d'un nouvel appareil et l'importance diagnostique de la méthode. *Acta Otorhinolaryng Belg* 16 5.
- Miyoshi, T. 1970. Analysis of optokinetic nystagmus using electronic computer. *Pract Otol Kyoto* 63 89.
- Monnier, M. 1967. Central mechanisms of vestibular and optokinetic nystagmus. *Ciba Found. Symp on Myotonic Kinesitherapy and Vest. Mech.* pp. 205-213.
- Monnier, M., Bellin, E. & Polc, P. 1970. Facilitation, inhibition and habituation of the vestibular responses. *Adv Oto-Rhino-Laryng* 17 25.
- Montandon, A., Hugonin, S. & Rohr, A. 1970. Vestibular habituation. Experimental ENG results. *Adv Oto-Rhino-Laryng* 17 129.
- Motohashi, H. 1965. Studies on the directional preponderance of nystagmus. *Jap J Otol Tokyo* 63 13.
- Oka, K. 1954. On the optokinetic training and nystagmus. *Pract Otol Kyoto* 47 896 (in Japanese).
- Pfaltz, C. R. & Piffko, P. 1970. Studies on habituation of the human vestibular system. *Adv Oto-Rhino-Laryng* 17 169.
- 1970. Retention of vestibular habituation. In *Vestibular function on earth and in space* (ed. J. Stahl), Werner-Gren Symposium no. 15 Pergamon Press, Oxford New York.

Pfakz, C. R. & Richter, H. R. 1955 La photo-électro-nystagmographie. Une nouvelle méthode d'enregistrement du nystagmus. *Compte Rend. Acad. Sci. Paris* 251 113.

— 1965 Photoelektrische Nystagmusregistrierung. *Pract. Otorhinolaryng.* (Basel) 18 263.

Palouy, P., Gilbert, J., Blanc, P., Chouard, Ch. & Fontelle, H. 1970. Study on vestibular habituation among pilots and flying staff in terms of their training and seniority. *Adv. Oto-Rhino-Laryng.* 17 167.

Richter, H. R., Pfakz, C. R. & Haerri, W. 1966. La rapidité des mouvements oculaires et les conditions d'enregistrement en oculo- et nystagmographie. *Conf. Neurol.* 28 300.

Sharpless, S. & Jasper, H. 1956. Habituation of the arousal reaction. *Brain* 79 635.

Shiraki, T. 1967. Response increase of optokinetic nystagmus. *Pract. Otol. Kyoto* 60 970 (in Japanese).

Thorpe, W. H. 1950. The concepts of learning and their relations to those of instinct. In *Physiological mechanisms in animal behaviour* (ed. Danieli & Brown). Cambridge Univ. Press.

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DISCUSSION

M. Hinoi: We too have studied the mechanism of transfer of the effects of repeated optokinetic stimulation to labyrinthine nystagmus. As pointed out by Dr Oka, in Oka, Japan, when guinea pigs received repeated optokinetic stimulation they showed an increased response in optokinetic eye nystagmus. In parallel with this, they developed a response decline in postrotatory eye nystagmus. We confirmed this type of transfer by using rabbits and fowls. We also found that in rabbits in which the deep neck muscles were paralysed with procaine, this type of transfer was not observed appreciably even when adequate optokinetic stimulation was given repeatedly. This finding indicates that transfer of the effects of repeated optokinetic stimulation to labyrinthine nystagmus can only develop clearly when the cervical proprioceptors are functioning normally. In our experiments using EEG we also found that procainization of the deep neck muscles can greatly depress the activity of the activity of the central nervous system and especially the brain stem reticular formation. So, we may conclude from these findings that the brain stem reticular formation can play an important role in the mechanism of transfer of the effects of repeated optokinetic stimulation to nystagmus responses of labyrinthine origin.

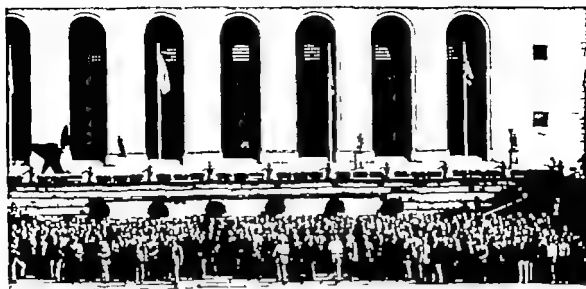


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THE XVIII CONGRESS OF THE SCANDINAVIAN OTO-LARYNGOLOGICAL SOCIETY

Göteborg, Sweden, June 28-30, 1972

The Congress was held in Stenhammarsalen, the Concert Hall Göteborg. The program was limited to the following topics.

Pathogenesis and Therapy of Chronic Otitis
Carcinoma of the Oral Cavity
Noise
Traumatology in Otology
Recurrent Nerve Paralysis
Prolonged Endotracheal Intubation.

The President, Professor G. Herberts, addressed the Congress as follows:

On behalf of the Congress Board I have the honour to wish all the members of the Congress from the different Scandinavian countries a hearty welcome to Göteborg.

The programs from earlier meetings during the activity of the Scandinavian Society have reflected the actual research fields of their day and the development of the speciality I wish

to remind you that at the first meeting of our Society now sixty years ago the famous Danish otologist Ernst Schmiegelow lectured on the treatment of sequelae of larynx and trachea after tracheostomy while we at this meeting are going to discuss a similar subject, namely lesions after endotracheal intubation instead of tracheostomy. We shall also be discussing other oto-laryngological themes, which have been selected after an inquiry among the colleagues. In this way we hope to receive an exposé of the problems of today in our speciality.

In the lay-out of the transactions we have tried to pay due attention to expressed wishes and we have also had the ambition to make the various subjects informative and responsive. To realize this, it has been necessary to increase the co-ordination of the lectures and arrangements in symposia and panel discussions, modelled by the respective moderator

As in earlier congresses several scientific exhibitions have been arranged to illustrate the various sections.

Collaboration in Scandinavian Otolaryngology is to be discussed at today's business meeting, especially some questions concerning organization and education at different levels. This is of especial interest before the expected reorganisation of the training programs.

It is our hope that the Congress will give a good presentation of the problems and the situation of otology today and make valuable contributions to the development in some important fields. In this way the tradition will be supported that was formed by the great predecessors in Scandinavian otology at the beginning of the century at the time when our Society was founded.

With these words I wish the members welcome to the transactions which will start immediately

Business meeting of the Congress

The President announced the Icelandic invitation for the next Congress to be held in Reykjavik in 1975. The Congress accepted the invitation unanimously and agreed to the proposal that the next Congress Committee should comprise Dr E. Thorsteinsson, President Dr

S. Skaftasson Vice President and Dr Ole Bentzen General Secretary. It was suggested that the Congress should take place on June 29 to July 2, but decisions upon date and program were left to this Congress Committee.

The Icelandic members asked the members of the other Scandinavian countries to support them in the development of oto-rhino-laryngology in Iceland. The Congress recommended the Otolaryngologic Societies and Associations of the different countries to give the Icelandic members all information and support which they might need.

Professor H. Engström proposed that educational and examination material at all levels in oto-rhino-laryngology should be exchanged between the different university clinics and other clinics involved in education in the Scandinavian countries. The Congress accepted this proposal and the Department of Otolaryngology in Uppsala was chosen to act as a bank for this material.

Professor H. C. Andersen recommended a meeting to discuss education at all levels in oto-rhino-laryngology in the Scandinavian countries. The Congress accepted the idea and Professor Andersen promised to prepare proposals for such a meeting.

NEW ASPECTS IN THE PATHOGENESIS OF CHRONIC SECRETORY OTITIS MEDIA

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Abstract On the basis of quantitative studies of the mucous elements in the Eustachian tube and middle ear mucosa of normal and abnormal temporal bones as well as of biopsies from secretory and adhesive otitis the secretory pathogenesis is described. Secretory otitis is divided into a stage of development, during which the mucous glands form, a stage of secretion, during which they produce mucus, and a stage of degeneration, during which they degenerate, become inactive, and mucus production decreases. The relationship between the ability of the mucosa to produce mucus and the ability of the Eustachian tube to transport the mucus to the rhinopharynx is the essential factor in the secretory pathogenesis.

The "transudation" theory or the "exudation theory" on the pathogenesis of chronic secretory otitis cannot explain why secretion in the middle ear of children is tenacious mucus which, despite a well-functioning drain, may form again, and why secretory otitis may be present even though there is perforation of the drum and a patent Eustachian tube. We have previously on the basis of quantitative studies of mucous glands in an autopsy specimen, advanced a new "secretory" theory on the pathogenesis of the disease (Tos & Bak Pedersen 1972). Continued quantitative studies of the mucous elements in normal and abnormal autopsy specimens and in biopsies from secretory and adhesive otitis have disclosed new findings which confirm and supplement this theory on the pathogenesis. Foetuses have no mucous glands in the osseous tube or in the middle ear. Children and adults whose

tubal or middle-ear mucosa has never been exposed to abnormal action show only a few glands or none, but a larger number if the mucosa has been exposed to an abnormal action. Since most of the glands become inactive, such ears may be clinically quite normal.

In an autopsy specimen with mild chronic secretory otitis we found 488 mucous glands in different secretory stages (Bak Pedersen & Tos, 1971) and a marked increase of goblet cell density. In biopsies from 85 ears with typical secretory otitis and mucus in the middle ear we found mucous glands in all but two (Tos & Bak Pedersen, 1972). The mean density was 71 glands/mm² corresponding to about 2 000 glands in the entire middle ear 93% of which were active. The goblet-cell density was greatly increased. In biopsies from 48 ears with chronic adhesive otitis we found a density of 4.3 glands/mm² only 11% of which were active.

The types of glands show given characteristics. A distinct transition from active to degenerated glands was demonstrated. Active glands have well-preserved secretory epithelium, narrow ducts, and no stagnation of mucus in the tubules. Transitional varieties have a somewhat dilated duct system filled with mucus, but the secretory epithelium is still fairly well-preserved in places. Degenerated glands show maximum dilatation, a mucus-filled duct system where the secretory

epithelium has entirely disappeared. The primary factor in degeneration of glands seems to be stasis of mucus flow causing dilatation of the duct system, stretching of and pressure upon the secretory epithelium with degeneration and total disappearance of the mucous cells.

Abnormal glands must form at some stage of secretory otitis, which we call the stage of development. The histology of this stage has not yet been described, but in biopsies we found some developing young glands. The glands form a link in epithelial metaplasia due to low virulent infection: chronic catarrhal state of the tube, long-lasting tubal occlusion or recurrent otitis. The irritant is presumably chronic or recurrent. At least 1-2 months have to pass before the glands have completed their development. When they form mucus, the stage of secretion starts, mucus accumulates in the middle ear and causes the well known clinical symptoms. Histology is predominated by active mucous glands and highly increased goblet-cell density. The period elapsing until the middle ear has filled with mucus depends upon the secretory capacity (number and size) as well as secretory activity of the glands and not least upon tubal and ciliary function which transports the produced mucus to the rhinopharynx. At a slight secretory capacity and good tubal function the produced mucus can be transported to the rhinopharynx, and major accumulations will never occur in the middle ear. Conversely with a large number of active glands and poor tubal function the middle ear soon fills, and despite a well functioning drain mucus will regenerate in the course of a month or two. The relationship between the ability of the mucosa to produce mucus and the ability of the tube to transport it away from the middle ear is the essential factor in the secretory pathogenesis and explains the varying degrees of severity and duration of the disease. However the aetiological factors may cause renewed formation of glands and increased secretory capacity. If untreated, the disease enters a vicious circle. Accumulation

of mucus, exacerbation of tubal occlusion, formation of new glands, and increased mucus production. When the glands start degenerating the disease enters the stage of degeneration in which the histology is predominated by the transitional varieties and degenerated glands and during which mucus production decreases. If tubal function returns to normal, the ear recovers, if adhesive otitis has not arisen already. The small amounts of mucus produced during this stage may be transported to the rhinopharynx through a well-functioning tube. However in this stage too new glands may form and the mucus production may increase. This explains why the disease may last for a very long time and flare up anew.

Quantitative studies of the mucous elements performed so far have shown that the secretory capacity of each gland is so slight that a few active and many degenerate glands in the middle ear are of no clinical importance. At least 200 active glands are required to cause accumulation of mucus. Goblet-cell density also increases during the course of the disease and decreases during the stage of degeneration.

The secretory pathogenesis fully explains the clinical course but not the presence of straw coloured fluid occasionally found in adults. Perhaps there is—unlike the condition in children—a special, more exudative form of the disease.

REFERENCES

- Bak Pedersen, K. & Tos, M. 1971. The mucous gland density in chronic secretory otitis media. *Acta Otolaryng (Stockh.)* 72: 14.
- Tos, M. & Bak Pedersen, K. 1972. The pathogenesis of chronic secretory otitis media. *Arch Otolaryng (Chic.)* 95: 511.
- 1972. Density of mucous glands in a biopsy-material in chronic secretory otitis media. *Acta Otolaryng (Stockh.)* 73: 55.

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MIDDLE EAR EPITHELIUM IN CHRONIC EAR DISEASE

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Abstract. Thirteen chronically infected temporal bones were serially sectioned and stained to study the characteristics of any possible squamous epithelium. Non-cholesteatomatous epithelium is either immigrative or metaplastic in origin and the cholesteatomas always immigrative. The metaplastic epithelium does not keratinize and the non-cholesteatomatous immigrative slightly keratinizing type may change into cholesteatomatous aggressive type only in the presence of ill present unknown factors.

The purpose of this paper was to examine systematically in serial sections the middle ear epithelium in chronically infected temporal bones, to study the characteristics and origin of the squamous epithelium, and to relate the epithelial changes to tympanic pathology in general. The material consists of 13 temporal bones, fixed in 10% formalin and decalcified with 22.5% formic acid-sodium citrate solution. After celloidin-paraffin embedding, the blocks were sectioned and stained by various methods for general characteristics, for mucosal secretory components and for epithelial phospholipids (Karma, 1972).

The middle ear epithelium in cases of chronic infection showed hypertrophy hyperplasia, secretory proliferation and squamous metaplasia. Immigrating squamous epithelium, epidermization and even epidermosis were also encountered in this material. Sometimes the opposite was the case: the tympanic epithelium emigrated to the outer surface of the drum membrane.

As a rule respiratory often secreting epithelium lined the middle ear cavity widely in its anterior and inferior parts, extending in many

ears to the inner surface of the tympanic membrane, to the epitympanum, posterior tympanum, and even to the area of the mastoid air cell system. The tubotympanic ciliated epithelium continued posteriorly in the form of one to four narrowing strips which were located mainly inferiorly and extended in some ears up to the posterior limits of the middle ear cleft.

Metaplastic columnar and squamous epithelium in various stages occurred in all ears; however the metaplastic stratified squamous epithelium never showed any signs of keratinization.

All epidermal layers of the immigrating stratified squamous epithelium, except rete pegs, were seen at least in the area of the perforation edge. The junction between the epidermis covering the tympanic membrane remnants and the tympanic epithelium varied from very abrupt change to a gradual change with a transitional area of varying length. In some ears the junction showed keratinizing squamous epithelium and columnar ciliated epithelium side by side and sometimes the squamous epithelium lost its keratinizing properties before it joined the tympanic columnar epithelium. In other ears the squamous epithelium grew for a short distance under the columnar epithelium and pushed it aside. In some cases it also grew for some tenths of a millimetre over the columnar epithelium. In 6 temporal bones the epidermis sent extensions deeper into the subepithelial area near the junction.

In cases with gradual changes the transitional epithelium had a length of 0.1 to 2 mm in the junctional area. The transition often occurred gradually through various parakeratotic and pseudoparakeratotic stages, first into metaplastic and later into columnar and cuboidal epithelium. The epidermal immigration was of the least extent in central perforations whereas total pars tensa defects showed wide immigration into the tympanum and to the promontory. The epithelium covering the promontory was squamous in 6 cases in 3 it was clearly immigrative and in 3 metaplastic.

The immigrating squamous epithelium retained its slight keratinizing properties in the tympanum and changed, in only one case into aggressive-looking hyperkeratotic and acanthotic epidermoid epithelium showing subepithelial extensions. Except hyperkeratosis there were no other definite histological differences between these two types of squamous epithelium.

It is obvious from these studies of serially sectioned temporal bones that in chronically infected middle ears the mucosa almost always shows areas of squamous epithelium of varying extent. It may be immigrating, less often metaplastic and it can retain its non-cholesteatomatous character throughout the duration of life.

Transition from ciliated into squamous epithelium and vice versa, and large differences in

keratinization have been described in experimental studies (McLoughlin, 1961). It is possible that mesenchymal factors play an important part in epidermal differentiation and keratinization.

These data confirm the findings of Palva et al. (1968) that there exist various types of squamous epithelium in chronically infected middle ears. Serial sections suggest that the non-cholesteatomatous type of squamous epithelium is either immigrating or metaplastic in origin and that the cholesteatomatous type is always immigrative. The metaplastic type does not undergo keratinization the non-cholesteatomatous immigrative type may retain its properties of slight keratinization permanently and does not change into cholesteatomatous aggressive, expansive epithelium, a process which is still poorly understood at present.

REFERENCES

- Karma, P. 1972. Middle ear epithelium and chronic ear disease. *Acta Otolaryng* (Stockh.), Suppl. 367.
 McLoughlin, C. B. 1961. The importance of mesenchymal factors in the differentiation of chick epidermis. *J Embryol Exp Morphol* 9: 385.
 Palva, T., Palva, A. & Dammert, A. 1968. Middle ear mucosa and chronic ear disease. *Arch Otolaryng* (Chic.) 87: 21.

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DENSITY OF MUCOUS GLANDS IN VARIOUS CHRONIC MIDDLE EAR DISEASES

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Abstract From 491 operated ears 5180 pieces of mucosa from 16 different middle-ear sites were stained by the PAS-alcan blue whole-mount method and the density of glands in each piece determined. The mean density of glands was greatest in secretory otitis, 7.1 glands/mm² in dry adhesive otitis, 4.3 in chronic, infected, adhesive otitis, 3.4 in chronic granulating otitis, 2.0 in cholesteatoma and sequestrae to otitis, 1.3 in tympanosclerosis, 0.6, and in otosclerosis, 1.7. The differences in glandular density and the predominance of the various secretory types were found to be characteristic of the various middle-ear diseases.

Research into the histopathology of the human middle-ear mucosa in various chronic middle-ear diseases has increased during recent years. This research has been based partly on biopsies (Bendek, 1963; Sadé & Wernberg, 1969) and partly upon post-mortem studies of temporal bones (Sadé, 1966; Bak Pedersen & Tos, 1971) and has been concerned *in al.* with the mucous elements of the middle-ear mucosa—glands and goblet cells—which have been demonstrated by most authors. However apart from Bak Pedersen & Tos study from 1971 the investigations conducted so far have been on sections which do not permit an exact quantitative determination of the mucous elements. The present study was carried out by a special whole-mount technique which makes it possible to determine the density of the mucous glands, expressed as glands/mm² in the various middle-ear diseases as well as a more accurate morphological assessment of

the individual glands. Such quantitative studies have not been published previously.

MATERIAL AND METHOD

The material comprises 491 ears (Table I). The investigated pieces of mucosa were removed in various otosurgical procedures from 16 different sites in the middle ear and from 1–10 localities per patient. The number of biopsies depended upon the extent of the operation, the study being restricted to mucosa which was to be removed anyway. A total of 5180 mucosal pieces were studied. The tissue pieces were stained by the PAS-alcan blue whole-mount method (Tos, 1970). Under the stereomicroscope the mucous glands were counted, the area of the tissue pieces measured, and the density (glands/mm²) calculated, for each locality as well as for each ear.

RESULT

For all ears 6331 mm² mucosa was counted and showed 13325 glands. Of this number 20% were active glands with PAS-alcan blue positive secretory epithelium and narrow tubular lumina without stagnant mucus. 31% were large glands showing cystic degeneration with flat epithelium devoid of secretory activity marked dilatation of the tubules and excretory duct, as well as stagnation of mucus.

Table I The material studied

	No. of ears	No of studied sites	Total no. of glands	Total area (mm ²)	Mean density (glands/mm ²)
Secretory otitis	85	85	743	105	7.1
Dry adhesive otitis	48	151	1 855	424	4.3
Chronically infected adhesive otitis	30	105	2 539	752	3.4
Chronically granulating otitis	93	316	5 166	2 612	2.0
Cholesteatoma	70	241	1 982	1 510	1.3
Sequelae to otitis media (dry)	95	189	873	682	1.3
Tympanosclerosis of tymp. cav	26	47	60	184	0.6
Otosclerosis	44	56	107	63	1.7
Total	491	1 190	13 325	6 331	

49.6% were transitional varieties between the active and the degenerated glands, but showing degeneration of the greater part of the secretory epithelium. The ratio between active and degenerated glands in secretory otitis showed a great predominance of active glands (93%). In adhesive otitis, chronic granulating otitis, cholesteatoma, and sequelae to otitis the active glands made up only 8-20% in tympanosclerosis there were but few and in otosclerosis no active glands. The remainder were transitional varieties and degenerated glands.

The mean density of glands was greatest in chronic secretory otitis and least in tympanosclerosis (Table I). Within the individual disease groups there was quite considerable dispersion of the density partly in the different middle-ear sites, and partly between the different ears. The widest range was observed in adhesive otitis which showed in one site (annulus posteriorly) a range of from 0-68 glands/mm².

The density of glands, and especially the ratio between active and inactive glands, form

part of a given pattern of the histopathology of the middle-ear mucosa in the various middle-ear diseases. The secretory activity and the secretory capacity of the glands are of significance in the treatment and prognosis of the various middle-ear diseases.

REFERENCES

- Bak-Pedersen, K. & Tos, M. 1971. The mucous glands in chronic secretory otitis media. *Acta Otolaryng* (Stockh.) 72: 13.
- Bendel, G. A. 1963. Histopathology of transitory secretory otitis media. *Arch Otolaryng* (Chic.) 74: 33.
- Saddé, J. 1966. Pathology and pathogenesis of serous otitis media. *Arch Otolaryng* (Chic.) 84: 297.
- Saddé, J. & Weinberg, J. 1969. Mucus production in the chronically infected middle ear. *Ann Otol* 78: 148.
- Tos, M. 1970. Mucous glands of the trachea in children. Quantitative studies. *Annl Amz* 126: 145.

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CICATRICAL CHANGES OF THE EARDRUM AFTER TREATMENT WITH GROMMETS

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Abstract 79 of 117 patients, treated with grommets for chronic secretory otitis media, developed cicatricial changes (atrophy or/and fibrosis) of the ear drums. 11 of these patients had only minor changes at the former grommet site. Atrophic drum was related to a "negative" middle ear pressure (i.e. dysfunction of the Eustachian tube). Fibrosis of the drum, in some cases in combination with atrophy was found more frequently among patients with long drainage time. Hearing was not affected in any of the groups.

One of the problems in the treatment of chronic secretory otitis media by transmyringal drainage with plastic tube (grommet) is: does the treatment produce cicatricial changes of the eardrum and if so do these affect hearing? In a retrospective investigation with follow-up examination of patients treated with grommets, we have tried to explore this problem.

MATERIAL AND METHOD

The material includes 117 patients (188 ears) treated with grommets (i.e. tubulation) for chronic secretory otitis media. (In total 146 patients were treated, 29 absented themselves from follow-up and are not included in the material.)

Of the patients 103 were children and young teenagers (<15 years). The age distribution had concentration around 3-4 years (at the moment of tubulation). Two-thirds of the patients were males. Average observation time (i.e. time from extrusion of the grommet to follow-up) was 22 months.

The follow-up examination included otoscopy with classification of present cicatricial changes of the eardrums as listed in Table I (using the common name cicatricial for both atrophic and fibrotic changes). Furthermore audiometry, tympanometry by impedance bridge, examination of stapedius reflex and (usually) inspection and photography of the ear-drums by the operation microscope.

The data of right and left ear in the binaurally treated patients (71 persons) showed significant concordance. In order to avoid falsely significant results only one set of data were used from each patient (one of the ears selected at random).

RESULT

Seventy-nine of 117 patients developed cicatricial changes of the eardrums between the first examination at the clinic and follow-up (Table I).

By examining the dependence between the groups listed in Table I, and parameters considered by us to play a possible role in the development of the cicatricial changes, the following results were obtained.

The more severe cicatricial changes (groups 2-8) were found in all age groups with a frequency of 30-50%. Fibrosis dominated in patients treated at an age under 3 years, while atrophy was more frequent in the higher age groups.

Table I Classification and distribution of the cicatricial changes developed during or after tubulation

	No. of pats.
1 Minor changes at the former grommet site	38
2 Isolated small atrophic scar	3
3 Large atrophic scar	1
4 Atrophic drum	10
5 Slightly fibrotic drum	14
6 Severely fibrotic drum	1
7 Mixed fibrosis and atrophy slight degree	11
8 Mixed fibrosis and atrophy severe degree	1

Atrophy of the eardrum (group 4) In this group 80% of the patients had a "negative" middle ear pressure (less than -200 mm water) against average for all patients; 29%. Average drainage time was 5.8 months in the atrophy group against average for all patients; 6.3 months.

Fibrosis of the eardrum (groups 5-6) Middle ear pressure was "negative" in only 20% of the patients while average drainage time in this group was 7.3 months.

Mixed fibrosis and atrophy (groups 7-8) In this group middle ear pressure was found to be "negative" in 33% of the patients. Average drainage time was 9.0 months.

Anamnestic duration of symptoms, repeated tubulation, suppuration during drainage and sex showed no relation to type or degree of the cicatricial changes of the eardrum.

Hearing At follow-up was not affected in any of the larger groups (4, 5+6 and 7+8). An average hearing loss of 10 dB was found in each of the groups as well as in the whole material. The stapedius reflex could not be released in 80% of the patients in the atrophy group in 30% of the patients in the fibrosis

group and in 55% of the patients in the group with mixed fibrosis and atrophy.

DISCUSSION

Atrophy of the eardrum seems to be connected with a "negative" middle ear pressure, probably as a result of impaired function of the Eustachian tube. Average drainage time and number of tubulations in the group with atrophic drum did not exceed the average for the whole material, indicating that the grommet plays hardly any part in producing this type of change.

Our results suggest that development of fibrosis of the eardrum may be a side effect of treatment with grommets. This conclusion is drawn from the fact that average drainage time was longer in the group with fibrosis and the group with mixed fibrosis and atrophy than in the whole material. In the mixed group "horseshoe hyalinosis" was a characteristic finding: a thickened (hyalin fibrotic) part of the eardrum around an atrophic area, the former grommet site (Mackinnon, 1971).

In conclusion, treatment with grommets eliminates the "negative" middle ear pressure, but a risk of development of fibrosis of the eardrum may exist.

REFERENCES

- Mackinnon, D. M. 1971. The sequel to myringotomy for otodysplastic otitis media. *J. Laryng.* 85: 773.

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TYMPANOMAXILLARY SHUNT

Experiences and Development

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Abstract Various late complications have occurred in 6 patients following insertion of a tympanomaxillary shunt. The most common of these, slipping and displacement of the shunt, can probably be prevented by using a prefabricated curved tube, by fixating the sutures with silastic adhesive and by putting the tube outside the bony canal wall. This appears to be the only reported method of tubal by-pass which restores middle ear ventilation leaving the canal skin and ear drum intact and which allows the patient to auto-inflate by performing Valsalva's manoeuvre.

Tympanomaxillary shunt (TMS) was introduced in 1967 (Drettner & Ekvall, 1967 1969 1970) as a method of reestablishing the aeration of the middle ear in cases with chronic obstruction of the Eustachian tube. A silicone rubber tube is placed from the anterior part of the middle ear to the maxillary sinus, running above the temporomandibular joint and medial to the zygoma. The ear drum and the canal skin remain intact after the operation.

After insertion of the shunt the patient can maintain middle ear ventilation by performing Valsalva's manoeuvre which is not possible in patients treated by insertion of an indwelling tube through the drum.

Indication for TMS

Chronic obstruction of the Eustachian tube which has failed to respond to conventional methods of treatment.

Contraindications

Age (not before puberty).

Recurrent infections of the nose and/or para-

nasal sinuses (patients with a cleft lip and palate are thus usually not suitable for TMS). Hypoplasia of the maxillary sinus.

Radiotherapy (tumour doses to the operative area (e.g. for nasopharyngeal malignancies).

Active middle ear infection (e.g. discharging chronic otitis).

Material

To date the operation has been performed on 6 Swedish patients. Furthermore, a few similar operations have been performed in the United States (Glasscock, 1971 Joseph, 1971). The Swedish patients were aged 15 to 47 years at operation and the length of follow-up varied between 5 1/2 years and 1 year.

Results and complications

Restoration of middle ear ventilation together with hearing improvement was achieved in the early postoperative period in all patients. However 1 patient developed autophonia. This disappeared when the shunt, which was wider than in the other cases, was replaced by a narrower one.

Several complications have occurred in the late postoperative period. Slipping of the shunt in the ear canal has occurred in 3 patients. All these patients had a straight tube placed in a groove in the ear canal and bent at the zygomatic root at operation. After 1-5 years the tube became displaced and lost contact with the bony canal wall, perforating the canal skin. In 2 of these 3 patients the shunt was extruded through the tympanic membrane. These

This work was supported by the Swedish Medical Research Council (Proj. No. 17X 749).

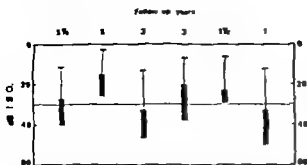


Fig. 1. Hearing results in 6 patients with tympano-maillary shunts. Pre- and postoperative hearing are given as means of the frequencies 500, 1000 and 2000 Hz. Filled bars indicate the hearing gain. Horizontal line indicates bone conduction threshold.

2 cases were revised, one with implantation of a new shunt and the other with removal of the shunt 3 years after the first operation. In the latter patient the middle ear was found to be aerated after removal of the shunt showing that while the TMS was in place the function of the Eustachian tube had recovered. The third patient with a displacement of the shunt has at present only a small area of the shunt which is uncovered by the canal skin, and as it is still in a satisfactory position in the middle ear no revision is indicated.

One patient developed recurrence of his adhesive otitis 6 months after insertion of the TMS. At revision the shunt was found to be patent but the silicone rubber sheet was displaced posteriorly.

A more serious complication was a long-lasting middle ear infection following acute sinusitis that occurred in one patient 6 months after insertion of the TMS. This patient was a case of ectodermal dysplasia with generalized dermatological manifestations and low resistance to infections. The TMS was removed.

At present the hearing level in all patients shows improvement (Fig. 1).

Technical improvements of the method

Early on in the development of the TMS technique it was realized that there was a risk of

late displacement of a straight tube placed in a groove of the ear canal and bent at the zygomatic root even if it was fixed by sutures. Before this risk became a reality a special curved tube was designed and is now manufactured (Extracorporeal Medical Specialties, King of Prussia, Pennsylvania, USA). This tube has an outer diameter of 2.5 mm and an inner of 1.2 mm. Silastic Medical Adhesive (Dow Corning Corporation, Midland, Michigan, USA) can also be used in conjunction with the sutures around the tube to prevent slipping of the sutures. Furthermore the surgical procedure has been modified in the more recent operations. The tube is now positioned in run from the anterior part of the middle ear into the pretympenic recess, through the anterior part of eptympanum up to the zygomatic root and it therefore mainly lies external to the preserved bony canal wall. These 3 steps (curved tube, silastic adhesive and surgical position of the TMS) will probably reduce the risk of displacement of the shunt.

The risk of middle ear infection cannot be reduced by these precautions. This stresses the contraindication of using a TMS in patients with a history of recurrent infections of the nose and/or paranasal sinuses.

REFERENCES

- Drettner, B. & Ekvall, L. 1967. En ny metod för återställande av mellanslans ventilation. *Nord Med* 78: 1594.
- 1969. Tympano-maillary shunt. A new method of middle ear ventilation. *Arch Otolaryng (Chic)* 90: 122.
- 1970. Chronic obstruction of Eustachian tube treated with tympano-maillary shunt. *Arch Otolaryng (Stockh)*, Suppl. 263: 29.
- Glascock III, M. E. 1971. Personal communication.
- Joseph, R. B. 1971. Personal communication.

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TOTAL MIDDLE EAR RECONSTRUCTION

L. Ekvall

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Abstract Surgical technique and postoperative results are reported in 56 cases of total middle ear reconstruction in classical radical cavities. In 93% of the cases a normal or nearly normal anatomy of the drum and the ear canal was achieved, permitting the patient to swim. 83% reached the 30 dB level postoperatively or closed the air-bone gap to within 10 dB. The results were better in cases with remnant mobile stapes than in cases without stapes superstructure.

Tympanoplasty in radical cavities involves 4 main problems:

1. A definitive clearing of the cavity and of the middle ear is a prerequisite of "closing" a previously open cavity
2. Reconstruction of a normal external auditory canal gives the patient a self-cleaning ear that will tolerate swimming, etc.
3. Restoration of ventilated spaces in the middle ear and in the mastoidectomy cavity is essential for the functional as well as for the anatomical result.
4. The sound transmission mechanism has to be restored.

METHOD

Through a retrosauricular incision the cortical opening of the cavity is exposed, care being taken not to injure any uncovered dura or sigmoid sinus. If possible the squamous epithelium of the cavity and of the middle ear should be dissected in continuity to avoid residual disease. Preoperative staining of the cavity with 2% gentian violet will facilitate

this step. The part of the meatal skin that turns posteriorly and superiorly inside the meatal orifice is resected to prevent postoperative skin pockets. Any remaining infected mastoid cells are exenterated. Care is taken to save as much as possible of the mucous membrane of the middle ear especially in the oval window niche and in the Eustachian tube orifice. Following cleaning, the cavity and the middle ear are lined with Silastic sheeting, which should reach down into the Eustachian tube orifice and posteriorly and superiorly cover the sigmoid and the dural plates. This will secure a large ventilated cleft from the tubal orifice to the periphery of the cavity and counteract postoperative retraction pockets. Through an aperture in the Silastic a 6-8 mm high "boomerang"-shaped columella, prepared from cortical bone or ossicular homograft, is placed on the centrally denuded stapes footplate. In cases with a fixed footplate and in most draining ears columellization is left for a second-stage operation. If the walls of the oval window niche are denuded of mucoperiosteum the long leg of the columella is partly covered with a 1.2 mm slitted Silastic tube to prevent bony fixation. In the case of

Table I

Healed drum and ear canal	51 (93 %)
Postoperative perforation	0
Postoperative cavity (intentional)	2
Postoperative retraction	3
	7 %

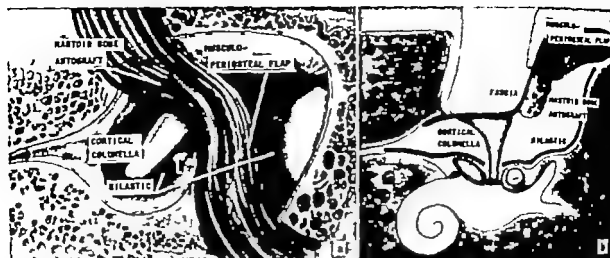


Fig. 1 (a, b) Schematic drawings to illustrate the surgical technique of total middle ear reconstruction in a classical radical cavity

a remaining mobile stapes a "boot"-shaped columella is used. The middle ear is filled with Gelfoam and dry temporalis fascia is used as tympanic membrane graft. The graft is placed medial to the annulus anteriorly and inferiorly. Posteriorly and superiorly the fascia reaches the remaining part of the external canal. The bony canal is completed with an autograft usually taken from the apex of the mastoid process. The graft is placed in grooves the remaining parts of the bony canal and is supported by a double-pediced musculo-periosteal flap which also bridges the gap between the bone graft and the meatal orifice. The canal is packed in two layers with strips of surgical rayon and synthetic fibre wad. The pack is left in place for 7 days and then cortisone-antibiotic ear drops are applied locally until epithelialization is completed. Postoperative Eustachian tube function is followed and is supported if necessary.

Table II

Serviceable hearing	> 10 dB ISO
or air-bone gap	< 10 dB
Total (n = 53)	41 (83 %)
Stapes (n = 17)	16 (94 %)
Footplate (n = 36)	28 (78 %)

RESULT

Anatomical and functional results in 56 consecutive cases with an observation period of between 3 years and $\frac{1}{2}$ year are shown in Table I and II. In two of the cases the mastoidectomy cavity was intentionally left open owing to very large and adhesive dehiscences

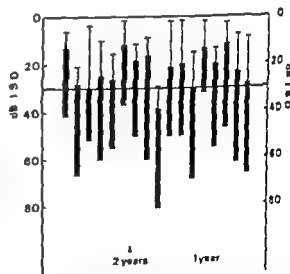


Fig. 2 () Individual hearing results in 17 cases with a remnant mobile stapes, ■ hearing improvement, □ hearing impairment. Postoperative bone conduction level is indicated by a horizontal line. The values are given as means of the speech frequencies.

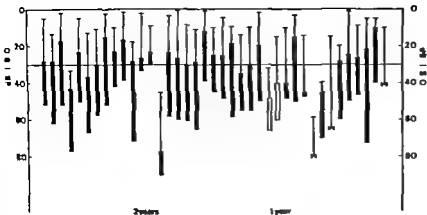


Fig. 2 (b) Individual hearing results in 36 cases with the stapes footplate as the only remaining ossicle. Caption as in Fig. 2 a. Δ ossiculoplasty revision; ∇ intentional second stage; \bullet only first stage performed.

of the facial nerve canal. Two cases have post operative retraction pockets with accumulation of squamous epithelium debris. Fifty-one ears have normal or nearly normal ear canal, permitting the patients to swim, etc. (Table I). In 53 cases an attempt to reconstruct the sound transmission mechanism has been made (Table II). In 3 cases this is planned at a second-stage operation. Six cases, all without stapes super structure, have undergone an ossiculoplasty revision via a tympanomental flap. Dislocation of the columella was found in 3 cases, secondary bony fixation of the footplate in 1 case, resorption of the long leg of the columella

in 1 case, and severe fibrous adhesions in 1 case. Table II shows the final hearing results so far. The less favourable results in the group with the stapes footplate as the only remaining ossicle, when compared with those in the other group, suggest that a mobile stapes from a functional point of view is the most valuable ossicle in tympanoplasty.

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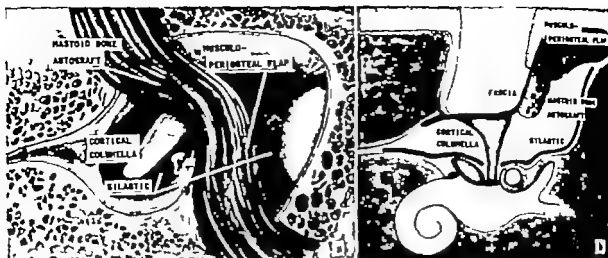


Fig 1 (a, b) Schematic drawings to illustrate the surgical technique of total middle ear reconstruction in a classical radical cavity

a remaining mobile stapes a "boot"-shaped columella is used. The middle ear is filled with Gelfoam and dry temporalis fascia is used as tympanic membrane graft. The graft is placed medial to the annulus anteriorly and inferiorly. Posteriorly and superiorly the fascia reaches the remaining part of the external canal. The bony canal is completed with an autograft usually taken from the apex of the mastoid process. The graft is placed in grooves on the remaining parts of the bony canal and is supported by a double-pediced musculo-periosteal flap which also bridges the gap between the bone graft and the meatal orifice. The canal is packed in two layers with strips of surgical rayon and synthetic fibre wad. The pack is left in place for 7 days and then cortisone-antibiotic ear drops are applied locally until epithelialization is completed. Postoperative Eustachian tube function is followed and is supported if necessary.

Table II

Serviceable hearing or air-bone gap	> 30 dB I.S.O. < 10 dB	
Total (n = 53)		44 (83 %)
Stapes (n = 17)		16 (94 %)
Footplate (n = 36)		28 (78 %)

RESULT

Anatomical and functional results in 56 consecutive cases with an observation period of between 3 years and $\frac{1}{2}$ year are shown in Table I and II. In two of the cases the mastoidectomy cavity was intentionally left open owing to very large and adhesive dehiscences.

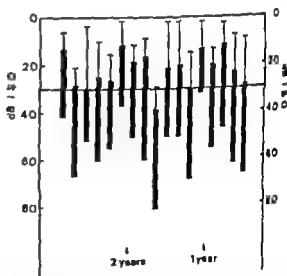


Fig 2 (a) Individual hearing results in 17 cases with a remanent mobile stapes. ■ hearing improvement; □, hearing impairment. Postoperative bone conduction level is indicated by a horizontal line. The values are given as means of the speech frequencies.

tween the drum and the stapedial plate the foot of the prosthesis was strengthened by means of a steel wire.

In control examinations it could be ascertained that the cartilage homograft heals excellently. In 6 cases the healing was disturbed by postoperative infection but this seldom occurred because of the reconstruction materials used. A large perforation was found in these cases shortly after the operation. Two ears were checked 16 and 20 months respectively after the primary surgery (myringoplasty) because of an unsuccessful hearing result, and in 15 ears out of 103 (14.6%) 3 perforations have subsequently healed spontaneously. The percentage is high but it should be kept in mind that ears with the largest perforations were chosen for cartilage myringoplasty. As parts of the sound conducting system the 8 grafts used have proved agreeable.

According to our experience the homoplastic septal cartilage is a serviceable alternative for use as reconstruction material of the eardrum, especially in cases of large or total perforation. The healing of the graft is good, and quite obviously the method really helps to retain the tympanic cavity in correct shape. It can be considered as an additional advantage

that no extra operation is necessary in order to obtain the reconstruction material. The cartilage was also found to be serviceable for the reconstruction of the ossicular chain, but there was no evidence of any superiority compared with, for instance incus autografts or homografts.

REFERENCES

- Brandow G 1970. Homograft tympanic membrane transplant in myringoplasty. *Acta Otorhinolaryng Belg* 24 38.
Gibson, T 1967. The transplantation of cartilage. *J Clin Path* 20 513.
James, C. 1963. Cartilage-tympanoplasty. *Laryngoscope* 73 1288.
— 1970. Homo- and heterogenous grafts in reconstruction of the sound conduction system. *Acta Otorhinolaryng Belg* 24 60.
Smyth, G & Kerr A. 1970. Cartilage homografts. Experimental and clinical aspects. *Acta Otorhinolaryng Belg* 24 53.
Venker J 1968. The restoration of the ossicular chain by means of cartilage. *Pract Otorhinolaryng (Basel)* 30 29.

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SECONDARY OPERATIONS FOR MIDDLE EAR INFLAMMATION

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Abstract Twelve per cent of operations for chronic middle ear inflammation during the period from May 1969 to Dec. 1971 were secondary operations. Among these 98 ears 33 recidivant of secondary acquired cholesteatomata were found. Several factors were considered to contribute to the need of secondary operations. Relevant preoperative treatment, meticulous operative technique and permanent follow-up are recommended as prerequisites of surgery for chronic middle ear cleft inflammation.

Of 1 206 middle ear operations performed during the period May 1969 to Dec. 1971 at the University Hospital of Turku 98 were secondary operations with an inflammatory middle ear disease as indication. The frequency of secondary operations was 12.1% taking into account only operations for chronic inflammation. Table I describes the patient series. In Table II the preoperative indication for the secondary operation is reported in relation to the findings at this secondary operation. Findings at the secondary operation confirmed by histopathological specimens are classified in five different types.

(1) Recidivant or secondary acquired cholesteatoma of the middle ear cleft.

(2) Chronic disturbing inflammatory reaction in the operatively created cavity of the middle ear.

(3) Recidivant inflammatory process in tympanic and/or mastoid region of the middle ear cleft.

(4) Scarring causing disturbances to the normal self-cleaning mechanism or significantly impaired hearing.

(5) Recidivant perforation of the drum.

Of the 98 secondary operations, 72 were second, 23 third, 1 fourth and one a fifth

operation for inflammation on the same ear. In all cases, a detailed analysis was made of circumstances and findings before the primary operation, at the primary operation and during the interval between the primary and the secondary operations. All this information was compared with findings at the secondary operation and the cases were fitted in one of the five types according to the above scheme.

As can be seen from Table II, in 16 cases the diagnosis of recidivant cholesteatoma was the main indication for the secondary operation. In 17 cases only continuing inflammation or disturbing scar formation requiring a reconstruction were the indication for the secondary operation which revealed a secondary cholesteatoma.

Main emphasis is paid to recidivant cholesteatoma as it represents a process that will always grow further and as such is often an absolute indication for operative treatment. Thirty three recidivant cholesteatomata were found among these secondary operations. Fourteen of these were considered to belong to the residual type, 12 to the recurrent or migration type and 7 to the iatrogenous type of cholesteatoma (Turner 1970). At the primary operation cholesteatoma was found in only 15 of 33 cases. In 5 exact information is not available and in 13 only chronic inflammation without cholesteatoma was revealed by the primary operation. When the type of primary operation was analysed in this group of secondary cholesteatomata, it was shown that any type of middle ear operation may be followed by secondary cholesteatoma. The recurrent or migration type of cholesteatoma is most often

Table I. Age distribution of patients at secondary operation

	Number of ears						
	<6	7-10	11-15	16-25	26-40	41-60	Totals
Female	2	6	11	10	13	9	51
Male	2	6	7	11	15	6	47
Total	4	12	18	21	28	15	98
	$\chi^2=1.52$ D.F. = 5						

found after simple mastoidectomy showing that the primary operation was not extensive enough in its type. The frequency of residual cholesteatoma was highest among modified radical operations with cavity obliteration technique. The incidence of intragenous cholesteatoma, which means an epithelial implant as a consequence of the operation, was almost equal among all the types of primary operation.

Comparison between recovery after the primary operation and findings at the secondary operation revealed that difficulties with inflammation are very common among patients of this series. Nevertheless there are 17 cases altogether in which no disturbance of healing could be seen after the primary operation. 3 of these cases belonged to the recidivant cholesteatoma group. An apparent technical failure in the exact fixing of the fascial transplant in relation to drum remnants was apparent in 13 cases, in 5 of which a cholesteatoma developed.

This type of examination cannot give a numerical estimate of the risk of secondary cholesteatoma or other secondary disturbance in connection with middle ear operations, as the primary operations of our series date from a period of more than 15 years. Our study revealed many factors that influence the appearance of secondary disturbance. Surprisingly little accumulation was seen for example in relation to primary operation for a dry ear or for an actively infected ear but again the manner of collection of this material prevents estimating any relative risk in this respect. In the light of this study we consider it always possible that secondary changes, either scar formation or cholesteatomatous degeneration, may occur even after a smooth healing process. The importance of a meticulous technique and careful preoperative treatment is stressed. A patient with chronic middle ear inflammation requires chronic follow-up even after apparently successful surgery. A careful follow-up is a prerequisite for surgery of inflammation in the middle ear.

REFERENCE

Turner J L. 1970. Cholesteatoma occurrence following miculoplasty obliteration of mastoidectomy cavities. *Laryngoscope* 80 1133

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Table II. Preoperative indications of secondary operation in relation to findings at this operation

Indications of secondary operation	Findings at secondary operation					Totals
	Cholesteatoma	Cavity inflammation	Mastoid-tympanic inflammation	Scar disturbance	Drum perforation	
Continuing inflammation	13	11	21	1	2	48
Secondary myringo-tympanic reconstruction	—	—	1	3	11	15
Ear canal or drum reconstruction	4	1	—	10	—	15
Recidivant or acquired cholesteatoma	16	—	—	—	—	16
Second stage of operation	—	—	—	4	—	4
Totals	33	12	22	18	13	98

SECONDARY OPERATIONS FOR MIDDLE EAR INFLAMMATION

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Abstract Twelve per cent of operations for chronic middle ear inflammation during the period from May 1969 to Dec. 1971 were secondary operations. Among these 98 ears 33 recidivant of secondary acquired cholesteatomata were found. Several factors were considered to contribute to the need of secondary operations. Relevant preoperative treatment, meticulous operative technique and permanent follow-up are recommended as prerequisites of surgery for chronic middle ear cleft inflammation.

Of 1 206 middle ear operations performed during the period May 1969 to Dec. 1971 at the University Hospital of Turku, 98 were secondary operations with an inflammatory middle ear disease as indication. The frequency of secondary operations was 12.1% taking into account only operations for chronic inflammation. Table I describes the patient series. In Table II the preoperative indication for the secondary operation is reported in relation to the findings at this secondary operation. Findings at the secondary operation confirmed by histopathological specimens are classified in five different types:

- (1) Recidivant or secondary acquired cholesteatoma of the middle ear cleft.
- (2) Chronic disturbing inflammatory reaction in the operatively created cavity of the middle ear
- (3) Recidivant inflammatory process in tympanic or/and mastoid region of the middle ear cleft.
- (4) Scarring causing disturbances to the normal self-cleaning mechanism or significantly impaired hearing
- (5) Recidivant perforation of the drum

Of the 98 secondary operations, 72 were second, 23 third, 2 fourth and one a fifth

operation for inflammation on the same ear. In all cases, a detailed analysis was made of circumstances and findings before the primary operation, at the primary operation and during the interval between the primary and the secondary operations. All this information was compared with findings at the secondary operation and the cases were fitted in one of the five types according to the above scheme.

As can be seen from Table II, in 16 cases the diagnosis of recidivant cholesteatoma was the main indication for the secondary operation. In 17 cases only continuing inflammation or disturbing scar formation requiring a reconstruction were the indication for the secondary operation which revealed a secondary cholesteatoma.

Main emphasis is paid to recidivant cholesteatoma, as it represents a process that will always grow further and as such is often an absolute indication for operative treatment. Thirty-three recidivant cholesteatomata were found among these secondary operations. Fourteen of these were considered to belong in the residual type, 12 to the recurrent or migration type and 7 to the iatrogenous type of cholesteatoma (Turner 1970). At the primary operation cholesteatoma was found in only 15 of 33 cases. In 5 exact information is not available and in 13 only chronic inflammation without cholesteatoma was revealed by the primary operation. When the type of primary operation was analysed in this group of secondary cholesteatomata, it was shown that any type of middle ear operation may be followed by secondary cholesteatoma. The recurrent or migration type of cholesteatoma is most often

Table II Comparison of results of tympanoplasty on 269 dry and 266 discharging ears assessed by the mean pre- and postoperative hearing the air-bone gap hearing gain, and TI

	Preop. hearing	Postop. hearing	Hearing gain	Preop air-bone gap	Postop. TI	Success ()
Dry	51.5	28.3	23.2	14.6	25.4	89
Discharging	50.9	36.7	14.7	23.0	33.3	69
Total series	51.2	33.0	19.0	19.1	29.3	79

radical cavities in 27% of the cases. However the mean hearing gain was most marked in old radical cavities (35.8 dB), least in sequelae to otitis (24.8 dB). In myringoplasty the mean hearing gain was 17.9 dB in type I tympanoplasty 21.0 dB in type II usually with interposition of the incus or cortical bone, 25.0 dB and in type III 24.1 dB in which we generally used as columella the incus as auto- or homo-graft as well as cortical bone. Type IV tympanoplasty was not performed on dry ears, in which a reconstruction of the middle ear to type III was always tried.

Discharging ears

All ears had had chronic discharge and could not be rendered dry by conservative measures. In 54 cases there was perforation of the pars flaccida, in the remaining cases of the tensa. 133 ears had a large or total perforation. The pathology of the ossicular chain especially of the stapes, was considerably more severe than in the dry ears (Table I). The main object was to clear the ear. In all 269 cases mastoidectomy was done, clearing all cells, removing the mucosa from the antrum and epitympanum, and removing the posterior ossicle mental wall. After radical removal of the cholesteatoma or granulations all ears had in the same stage tympanoplasty type I in 52, type II in 97 type III in 95 and type IV in only 22 cases. The cavity was obliterated by a bipartite, superiorly pedicled flap of the subcutis-muscles. The auditory canal was dilated.

The assessment of the results was as with dry ears. Prior to the operation 9% and after the operation 47% had social hearing. An air-bone gap of 0-15 dB was closed in 37% of

0-30 dB in 77% of the cases. Prior to the operation 69% had an air-bone gap greater than 30 dB, the remainder 15-30 dB. A hearing gain exceeding 10 dB was obtained in 66% of more than 20 dB in 32% 19% achieved a gain of 0-10 dB, 10% no gain, and 5% exacerbation of the hearing. Prior to the operation 16% had a TI of 30 dB or more after the operation 56% of the cases. The results were identical in cholesteatoma (130 cases) and in granulating otitis (72 cases). We were, therefore, not reserved in operating upon chronic granulating otitis and central perforation. The bony annulus (the bridge) was preserved and narrowed in 107 cases, removed in 159. The hearing gain was the same in both groups. Therefore, we do not plan to preserve the bridge at any price, especially if this decreases the chances of obtaining a dry ear. Eight ears have had constant discharge since the operation. The others have been dry at the completion of the treatment and for the following 6 months. The first part of the material was seen an average of 2 years after the operation 93% had dry ears. The results were most favourable in dry ears (Table II).

REFERENCES

- Tos, M. 1972. Assessment of the results of tympanoplasty. *J Laryng* 85 487
- 1972. Results of tympanoplasty with modified radical mastoidectomy. *Acta Otolaryng* (Stockh.) 74 61
- 1972. Tympanoplasty results and the age. *Arch Otolaryng* (Chic.) 96 493

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RESULTS OF TYMPANOPLASTY

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Abstract The results of tympanoplasty on 269 dry and on 266 ears with chronic discharge including mastoidectomy and tympanoplasty in the same stage, were assessed by different criteria and compared. The best results were obtained in dry ears, but those in discharging ears were not bad either. If the ear is not rendered dry by conservative measures, we recommend mastoidectomy and tympanoplasty in the same stage rather than in two stages.

There is still a surprising paucity of analyses on the results of studying major series assessing the entire material from a given period of time. From 1964 to 1970 we performed 535 tympanoplasties, 269 on dry and 266 on discharging ears. All ears operated on during this period were treated by tympanoplasty with six exceptions in which radical operation was carried out for special reasons. The results were assessed according to several criteria 3-9 months after the operation, in the range 500-2 000 cps.

Dry ears

Most of the ears had previously been discharging. 71 had been dry for 1-3 months, 76 for 3 months to 1 year and 122 for more than 1 year. In 146 cases the perforation was large or total, in 50 there was no perforation, but mainly incudal dislocations and adhesive otitis. In 103 cases the ossicular chain was intact but as a rule with tympanosclerotic, fibrous or osseous fixation in the remaining cases defective (Table I). Often the appearance of the attic, antrum or aditus was checked. Myringoplasty was carried out in 37 cases,

type I tympanoplasty in 70 type II in 116, and type III in 46.

Prior to the operation 7% and after the operation 67% had social hearing. An air-bone gap of 0-15 dB was obtained in 63% of 0-30 dB in 95% of the cases. Prior to the operation 75% of the cases had an air-bone gap exceeding 30 dB while 25% had 16-30 dB. A hearing gain of more than 10 dB was achieved in 87% of more than 20 dB in 56% of the cases. Hearing did not deteriorate in any of the cases, but in 3% it did not improve. Prior to the operation 14% and after 71% had a *TI* (threshold of intelligibility) of 30 dB or better. The results varied widely in the various disease conditions and depended highly upon the method of analysis. Thus, social hearing in sequelae to otitis was obtained in 82% in tympanosclerosis in 75% in adhesive otitis in 52% and in tympanoplasty on old

Table I Pathology of the ossicular chain in 269 dry and 266 discharging ears

	Dry	Discharging	Total
Intact	103	54	157
Lacking lenticular process	24	18	42
Lacking long process of the incus	88	134	222
Lacking long entire incus	15	47	62
Lacking parts of the stapes	16	14	30
Lacking entire stapes	76	87	163
Lacking parts of the malleus	31	63	94
Lacking entire malleus	8	19	27
Dislocation, fracture	22	11	33

Table II Comparison of results of tympanoplasty on 269 dry and 266 discharging ears assessed by the mean pre- and postoperative hearing the air-bone gap hearing gain, and TI

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REFERENCES

- Toe, M. 1972. Assessment of the results of tympanoplasty. *J Laryng* 86 487.
- 1972. Results of tympanoplasty with modified radical mastoidectomy. *Acta Otolaryng* (Stockh) 74 61.
- 1972. Tympanoplasty results and the age. *Arch Otolaryng* (Chic) 96 493.

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ADVANTAGES AND DISADVANTAGES OF CLOSED AND OPEN TECHNIQUES IN CHOLESTEATOMA

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Abstract. One's aim even in cholesteatoma cases should be a closed technique—when this can be done without too great a risk.

The aim of treatment of a chronic ear is to arrive at a result that anatomically and physiologically is as close to the normal ear as possible. In cases of non-cholesteatomatous chronic inflammation it is, in fact, actually possible to reach a result that is very near the ideal we seek to obtain. In cholesteatoma cases the problem is much more difficult. In these we have the following alternatives:

1. A small cholesteatoma-pocket, which is free and totally removable: in such case it is possible to use closed technique and obtain a good hearing result.

2. In cases where the whole cholesteatoma cannot be successfully removed we can judge the situation in the following way. If we use closed technique we take the risk of a recidive cholesteatoma. Is this dangerous? Apparently not if we have the patient under observation and she is prepared beforehand for a new impending operation. In order to solve the problem in this way the patient's consent must be obtained.

If we consider that the use of the closed method is impossible in a certain case, we should endeavour to obtain a dry ear with as good hearing as possible—an ear that implies no risk for the patient. A thorough operative eradication of all diseased structures, together with a proper antibiotic-chemotherapy combined with corticosteroids, usually results in cessation of the inflammation. In a dry ear cholesteatoma is also less active. Open mastoid cavities are to be avoided. There are several methods of filling these cavities.

The fistulas of the labyrinth are discussed by Professor H. C. Andersen. Such a fistula has a decisive influence upon the operative method in these cases. Generally these fistulas are left untouched—particularly if the other ear is deaf—and an open technique is resorted to. However experience has shown that a closed technique can be successfully used even in such cases.

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OPERATIVE TECHNIQUE IN MASTOID OBLITERATION

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Abstract. A mentally based postauricular musculo-periosteal flap is recommended for cavity obliteration. With preserved canal wall, it prevents spreading of infections to the mastoid cavity. In cases with removed posterior bony wall, a new rigid wall is reconstructed using cortical bone chips and bone dust between fascia and musculo-periosteal flap.

Simple mastoidectomy in its classical form consists of removal of the mastoid air cell system via a postauricular incision down to the dural and sigmoid sinus lamina and labyrinthine capsule, with preservation of the posterior bony ear canal wall. After healing, a deep postmastoidectomy depression can often be seen behind the ear while in some the mastoid cavity may become lined with endothelium and the depression may remain small.

During the 1950s we (Palva et al., 1959, 1964) saw many cases of mastoiditis complicating acute otitis media. After mastoidectomy and healing, later otitic infections in some cases spread rapidly from the middle ear to involve also the area of the mastoid cavity. To preclude such recurrent mastoiditis, I began to obliterate the mastoid antrum with a mentally based musculo-periosteal pedicle flap which was fixed over the antrum by gelatin sponge (Palva & Pulkkinen, 1959; Palva, 1962). This method proved effective in the prevention of recurrent mastoiditis and, in addition, minimized the size of the postauricular depression.

Having initially used temporal muscle fascia only for drum reconstruction, I began to use it together with this mentally based musculo-

periosteal flap for ear canal reconstruction in radical ear surgery (Palva, 1962, 1963). Until then the creation of open radical mastoidectomy cavities was the routine method, and as there were often difficulties in postoperative treatment, my aim was to get rid of the cavity entirely. Recently we have used, for posterior canal wall reconstruction, the soft tissues in combination with periosteum-containing large cortical bone chips, cemented in place with cortical bone dust. With this method a new bony posterior ear canal wall can be accomplished in 6-8 weeks.

The postauricular incision is made through the skin only which is then undermined posteriorly. The musculo-periosteal flap can be tailored to a very big or smaller size depending upon the size of the expected cavity. After cutting the flap superiorly posteriorly and inferiorly it is dissected free of bone but left attached to the meatus. The planum mastoideum and the ear canal are then exposed. Cortical bone chips with some periosteum attached are removed with a broad chisel and stored in saline. Using a large Stryker burr and continuous liberal irrigation with suction, the bone dust is then collected from the healthy dense cortical bone areas (Perkins, 1972). Ampicillin solution is added to the bone dust.

Having completed the mastoidectomy I proceed with swing door incisions in the soft ear canal wall and reflect the flaps superiorly and inferiorly (Palva et al., 1969). By this

method an excellent view into the middle ear and attic is obtained. If there is no cholesteatoma a small rim of the bony meatal wall is removed, as in stapedectomy to expose the incudo-stapedial joint. Working from both sides of the bridge, as suggested by Myers & Schlosser (1960), one can do a thorough clean-up. The mastoid cavity is obliterated by the meotally based musculo-periosteal flap there being no need for canal wall reconstruction, as the wall is intact.

For elimination of attic cholesteatoma the posterior ear canal wall should only be removed partly leaving most of its inferior portion intact. In cases with more extensive tympanic cholesteatoma most of the posterior ear canal wall must generally be removed. In these ears the anterior part of the fascia is used for drum repair the ear canal is filled with antiseptic gauze to retain its original size and the remaining major part of the fascia is lifted up to cover the posterior side of the ear canal skin. Periosteum-containing cortical bone slices are then placed posterior to the fascia starting from the level of the horizontal canal up to the level of the cortical bone. These broad bone chips are cemented in place by the bone dust. The musculo-periosteal flap is now placed into the cavity posterior to this bony reconstruction.

If necessary gelatin sponge artificial bone or more cortical bone chips and bone dust may be used to fill the possibly remaining cavity. The tamponade can be removed from the ear canal in 6-7 days and the ear needs weekly checking for a period of 3-4 weeks. After this time the ear canal is well stabilized.

In recent years much has been written on the preservation of the posterior bony ear canal wall and this seems to be the main object

of the operation according to some reports. The canal wall can and should in practice be preserved in all cases of chronic otitis without cholesteatoma. In cases with small attic cholesteatoma, removal of the canal wall may be limited. However in cases with large cholesteatoma the first duty of the surgeon is to remove the cholesteatoma epithelium entirely and, in doing so he should not hesitate to remove as much of the posterior ear canal wall as is necessary for total eradication. The recurrence rate is high if the main emphasis is placed on posterior bony wall preservation, but remains low if the surgeon aims primarily to remove the disease. Total removal of the bony posterior canal wall does not compromise the postoperative hearing level though the tympanic cavity may become slightly shallower than with preservation of an intact posterior wall.

REFERENCES

- Myers, D. & Schlosser, W. D. 1960. Anterior-posterior technique for the treatment of chronic otitis media and mastoiditis. *Laryngoscope* 70: 78.
 Palva, T. 1962. Mastoiditis in children. *Laryngoscope* 72: 353.
 — 1962. Reconstruction of ear canal in surgery for chronic ear. *Arch Otolaryng (Chic.)* 75: 329.
 — 1963. Surgery of chronic ear without cavity. *Arch Otolaryng (Chic.)* 77: 570.
 Palva, T., Friedmann, I. & Palva, A. 1964. Mastoiditis in children. *J Laryng* 74: 977.
 Palva, T., Palva, A. & Järvelä, J. 1969. Myringoplasty. *Ann Otol* 78: 1074.
 Palva, T. & Pötkinen, K. 1959. Mastoiditis. *J Laryng* 73: 573.
 Perkins, R. 1972. Personal Communication.

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ONE STAGE VS. TWO-STAGE PROCEDURE FOR CHOLESTEATOMA

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Abstract The otosurgeon has to face the choice between a one-stage vs. a two-stage operation while eradicating a cholesteatoma. Referring to selected examples his considerations are discussed.

The operative treatment for chronic otitis unites the classic objective of *eradicating* a focus with the modern ambition to *stimulate* the hearing function. Such an achievement requires the elimination of complex pathological processes linked to such cell types as epithelium and connective tissue of skin and tympanic membrane, mucosal lining and osseous material of the temporal bone and ossicles. Chronic otitis also implies a physically abnormal milieu for sound transmission and protection within the tympanic cavity. An imbalance exists between sound stimulation and the cochlea. Furthermore, one must bear in mind that an optimal middle ear function is related to the mastoid process and a patent Eustachian tube. Chronic otitis ends in cholesteatoma, osteitis, a change in the mucosal vitality and disproportion between its serous and mucous producing elements. Other effects of the infection are hyalinization, calcification or ossification. These are all factors to be dealt with, whether one applies an open or a closed technique, or if one realizes it during a one-stage or a two-stage operation.

For the sake of brevity we postulate that the eradication of the chronic process has been carried out: two questions now arise (1) Using a conventional open technique, how far to go

in a one stage operation only and how far to go provided the plan is to return for a second stage after say 6 months? (2) How is the challenge replied to by those using a closed technique like CAT? (combined approach tympanoplasty).

In the following, some simple examples are constructed and the decision to use 1 or 2 stages is weighed.

1 Preoperatively the chain is considered intact. A marginal perforation with cholesteatoma is recognized in the attic. The surgeon preferring an open technique decides to eradicate the process in the attic, and only eventually extend to a radical cavity but saving the bridge and hence preserving the suspension of both hammer and anvil. The defect in the membrane is closed with fascia. Thus the operation is completed in one stage. The CAT procedure includes a mastoidectomy and a dissection of the cholesteatoma out of the attic. It is all completed in one stage and the surgeon maintains that his method excludes the risk of establishing a postoperative retraction under the bridge. The hearing result should be the same.

2. A cholesteatoma is located in the attic and the antrum. The long process of the anvil is missing. Using a conventional open technique the surgeon completes in one stage but has to sacrifice the bridge. Optimal hearing is brought about while adapting fascia and/or the membrane to the stapedial head. The anvil may also be used as a link between the stapes

and the head of the hammer in a one-stage trial. Alternatively the cholesteatoma is removed in a first step, and when the soft tissues are consolidated above the oval window the surgeon returns for the second stage some 3-6 months later. To ensure a maximum of sound energy transmission a lever system is constructed between stapes and the membrane. The CAT-surgeon may maintain that his one stage operation is the superior in saving the bony meatus wall, producing the best hearing and avoiding a persisting radical cavity.

3 The cholesteatoma is spreading to the attic and the mastoid process. The anvil is missing and the stapes plate is left. Following an open technique the cholesteatoma is removed during the first stage checking the facial nerve and the windows. Care is taken not to dislodge the plate in the presence of infection. The mucosal lining appears abnormal and the Eustachian tube may be partially blocked. During this first stage no attempt is made to reconstruct any columella effect. The closing endeavour with a good fascial transplant prepares the basis for a future sterile tympanic cavity in which the second stage is performed later. The surgeon who believes in a CAT procedure will do his operation in one stage, placing some kind of a columellar system on the plate. His opponents will argue against an attempt to complete in one stage and strongly recommend the acoustic repair to follow in the second, for fear of damage to the stapes plate during the first healing period. In situations with tympano-sclerosis of the oval window however the CAT-surgeon will take no steps to eliminate the sclerosis as part of a one-stage operation. He is determined to leave such sclerosis untouched and to return for a second

stage when the cavum tympani is deemed sterile.

4 The patient is under 10 years of age. Cholesteatoma is present and the hearing is very good. In children the expansive growth of cholesteatoma is rapid into the mastoid process, extending to the window niches. A good preoperative hearing is often maintained while the cholesteatoma acts as a false link. Recurrence is frequently encountered following both techniques, whether of 1 or 2 stages. The oval window niche is difficult to clear and so also is the tympanic and facial sinuses. The CAT-surgeon thoroughly eradicates the cholesteatoma in his first stage, avoiding any hazards in the stapes area. The restoration of hearing is postponed for the second operation when after a long observation no cholesteatoma recurrence is to be feared. When dealing with a child in a similar situation a two-stage operation is generally recommended. The consequence of an open method is the inconvenience of a radical cavity whereas the closed method has no drawbacks of this kind. Which method has the highest rate of recurrence? Apparently the closed one. Which method gives the best hearing? The advantages of the closed way are notable. It is up to the surgeon to select a suitable procedure guided by the prevailing circumstances and aided by the components of the middle ear still serviceable. By no means should a desire to solve all problems in one single stage be given priority when a two-stage operation is the safer in respect of preservation of the inner ear.

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HISTOLOGICAL MALIGNANCY GRADING OF SQUAMOUS CELL CARCINOMA OF THE PALATE

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Abstract The histological point system grading of malignancy applied in the present study to squamous cell carcinoma of the palate gives a considerably more accurate measure of the ultimate prognosis than the earlier used system solely based upon an estimation of the degree of the differentiation of the tumours. It seems justified to consider this type of histological evaluation as an improved basis for the choice of therapy in individual cases.

The prognostic information which can be obtained from the histological differentiation of squamous cell carcinomas in highly and poorly differentiated tumours has been demonstrated by Eneroth et al. (1972) in a study of palate tumours (Table I). Thus, 51% of the patients with highly differentiated carcinomas died of the tumour disease, compared with

Table I. Squamous cell carcinomas of the palate: prognosis solely in relation to the degree of differentiation of the tumours*

Histological features	No. of patients	Died of tumour disease	
		n	%
Highly differentiated	75	38	51
Poorly differentiated	57	25	44

*From Eneroth et al., 1972.

68% of the patients with poorly differentiated carcinomas. As was pointed out by Eneroth et al. (1972) the need for an improved morphological grading of malignancy is obvious.

Table II. Histologic grading of malignancy based upon the tumour cell population

	Points			
	1	2	3	4
Structure	Papillary and solid cords	Strands	Small cords and groups of cells	Marked cellular dissociation
Differentiation	Highly keratinization	Moderately some keratin.	Poorly minim. keratin.	Poorly no keratin.
Nuclear polymorphism	Few enlarged nuclei	Moderate number of enlarg. nuclei	Numerous irregular enlarg. nuclei	Anapl. immature enlarg. nuclei
Mitoses	Single	Moderate number	Great number	Numerous

Table III *Histologic grading of malignancy based upon the tumour-host relationship*

	Points			
	1	2	3	4
Mode of invasion	Well defined borderline	Cords, less marked border line	Groups of cells; no distinct borderline	Diffuse growth
Stage of invasion	Possibly	Micro-carc. (few cords)	Nodular into connect. tissue	Massive
Vascular invasion	None	Possibly	Few obvious	Numerous
Cellular response (plasmolymphocytic)	Marked	Moderate	Slight	None

Therefore we have used a new system for the histological evaluation of the grade of malignancy in squamous cell carcinoma of the palate. The system—also applied by us in evaluating the degree of malignancy of squamous cell carcinoma of the larynx (Jakobsen et al.)—is based upon a registering of 8 different morphological criteria in a 1-4 point scale giving total point values for the tumours from 8 to 32. In Table II is the tumour cell population and in Table III the

relation of the tumour to adjacent tissues registered according to the 1-4 point scale.

Present series

In the present study we have applied this point scale system for grading the malignancy of 110 cases of squamous cell carcinomas of the palate which have been followed up for at least 5 years (5-40 years). The incidence of death in the tumour disease has been used as the criterium of prognosis.

The histological analyses were performed without any knowledge of the clinical stage, the treatment, or the further course of the disease.

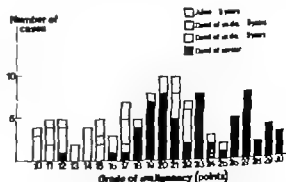
The results are presented in Fig. 1. It is obvious from the figure that a fairly good correlation exists between the histological grade of malignancy and the number of patients who died of the tumour disease. Thus, few patients

Table IV *Squamous cell carcinomas of the palate—survival time for patients who died of tumour disease*

Total	of patients Died of tumour	Time for death after treatment (within years)		
		1	2	3
110	62	44	55	59

Table V *Squamous cell carcinomas of the palate prognosis based on histological grades of malignancy*

Grades of malignancy points	No. of patients	Died of tumour disease	
		n	
10-16	28	2	7
17-23	55	34	64
24-30	27	25	93
Total	110	62	56

Fig. 1 *Histological malignancy grading of squamous cell carcinomas of the palate in relation to prognosis.*

died of tumours receiving a low point value, whereas patients with tumours acquiring a high point count run a great risk of dying from their tumour disease independent of other factors such as clinical stage and therapy. As is also seen from the Fig. 1 a fairly large number of patients had died of intercurrent disease within or after 5 years of follow-up. However the great majority of the patients who died of intercurrent disease after 5 years must be considered cured of their tumour disease, since out of 62 patients who died from tumour disease only 3 died later than 3 years after treatment (Table IV).

In order to make a direct comparison possible between the prognosis which was previously based upon the degree of differentiation of the tumours (Table I) and the prognosis which can be estimated from the point system applied, the material has been divided into 3 groups, each with a 7 point interval. In Table V the result of this follow-up study

is presented. Among patients with tumours acquiring a total point value between 10-16 only 7% have died from tumour disease. In the group with the high point value (24-30) no less than 93% of the patients have died of the tumour disease. The death rate from the tumour disease was also considerable in the intermediate group (17-23 points) 64% died from their tumours in this group.

REFERENCES

- Enneroth, C. M., Hjertman, L. & Moberger G. 1972. Squamous cell carcinomas of the palate. *Acta Otolaryng* (Stockh.) 73 418.
 Jakobsson, P. A., Enneroth, C. M., Kilander D., Moberger G. & Mårtensson, B. Histological classification and grading of malignancy in cancer of the larynx. *Acta Radiol* (in press).

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Table III *Histologic grading of malignancy based upon the tumour-host relationship*

	Points			
	1	2	3	4
Mode of invasion	Well defined borderline	Cords, less marked border line	Groups of cells; no distinct borderline	Diffuse growth
Stages of invasion	Possibly	Mikro-carc. (few cords)	Nodular into connect. tissue	Massive
Vascular invasion	None	Possibly	Few obvious	Numerous
Cellular response (plasmolymphocytic)	Marked	Moderate	Slight	None

Therefore we have used a new system for the histological evaluation of the grade of malignancy in squamous cell carcinoma of the palate. The system—also applied by us in evaluating the degree of malignancy of squamous cell carcinoma of the larynx (Jakobson et al.)—is based upon a registering of 8 different morphological criteria in a 1-4 point scale giving total point values for the tumours from 8 to 32. In Table II is the tumour cell population and in Table III the

relation of the tumour to adjacent tissues registered according to the 1-4 point scale.

Present series

In the present study we have applied this point scale system for grading the malignancy of 110 cases of squamous cell carcinomas of the palate, which have been followed up for at least 5 years (5-40 years). The incidence of death in the tumour disease has been used as the criterium of prognosis.

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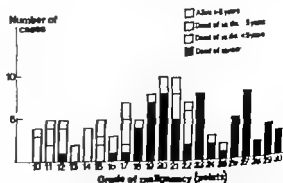
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REFERENCES

- Emeroth, C. M., Hjertqvist, L. & Moberger G. 1972. Squamous cell carcinomas of the palate. *Acta Otolaryng (Stockh)* 73: 418.
 Jakobsson, P. A., Emeroth, C. M., Kihlander D., Moberger G. & Mikrénsson, B. Histological classification and grading of malignancy in cancer of the larynx. *Acta Radiol* (In press).

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NUCLEAR DNA CONTENT AS A CRITERION OF MALIGNANCY IN SALIVARY GLAND TUMOURS OF THE ORAL CAVITY

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Abstract Deviation from the normal number of chromosomes is a characteristic feature of most malignant tumours and they are often great enough to cause changes in the nuclear content of DNA detectable by means of quantitative cytochemical methods. In malignant tumours of the parotid gland we have found that the nuclear DNA content of these tumour cells, as determined by quantitative cytophotometry was increased by 25 to 50% above the normal value. It was therefore believed that cytophotometric DNA analysis could add further information as to the question of malignancy of salivary gland tumours where morphologic methods are uncertain. Regarding the tumours of the oral cavity the question of malignancy is still a matter of controversy in two types of tumour namely monomorphic adenoma and highly differentiated mucoepidermoid tumour. The cytophotometric DNA analysis of the present study showed that the nuclei of the monomorphic adenoma

cells had identical DNA values with the normal cells. The highly differentiated mucoepidermoid tumour cells, on the other hand, exhibited higher DNA values, which were comparable with those of malignant parotid tumour cells. This adds further support to the idea that the monomorphic adenomas, despite the high cellularity are benign tumours, while the highly differentiated mucoepidermoid tumour should be considered as a malignant tumour.

During the last two decades various histological features of salivary gland tumours have been correlated with the corresponding clinical course in such a large number of patients that it has been possible to classify well delimited types of tumours as benign or malignant.

There are, however, two types of tumour in which the question of malignancy is still a

matter of controversy namely monomorphic adenoma and the highly differentiated type of mucoepidermoid tumour.

These two types are found mainly in the oral cavity where the relative incidence of mucoepidermoid tumour is about four times higher than in the major salivary glands (Eneroth, 1971). Monomorphic adenomas with a trabecular alveolar or tubular pattern are almost exclusively located in this region (Eneroth et al. 1972a). The high cellularity and the frequent lack of encapsulation of monomorphic adenomas (Eneroth et al. 1972a) are suspicious malignant morphological features. The morphologically benign structures, in cases with histologically highly differentiated mucoepidermoid tumour raise the question whether there is a benign variety of this type of tumour (Eneroth et al. 1972b).

Increased nuclear DNA content is generally considered as a characteristic feature of most malignant tumour cells and therefore an analysis of the nuclear DNA content has been made in order to add further information on the question of malignancy of these two types of tumour.

METHOD

After surgical removal of the tumours, imprint preparations were made and stained according to the Feulgen procedure. These

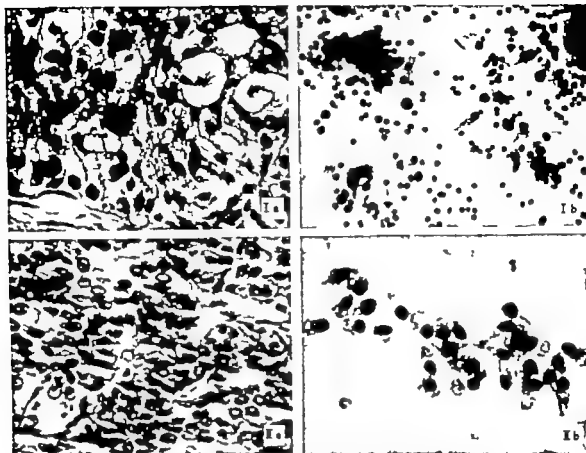


Fig. 1 I. Monomorphic adenoma of the palate. (a) Histological features. Photomicrograph, $\times 245$ (b) Imprints. Photomicrograph, $\times 245$ II. Highly differentiated

mucoepidermoid tumour of the palate. (a) Histological features. Photomicrograph, $\times 245$ (b) Imprints. Photomicrograph, $\times 245$

methods are described in detail elsewhere (Eneroth & Zetterberg, *in press*). Human lymphocytes from peripheral blood and cells from normal parotid glands were used as control cells. The DNA content of the Feulgen-stained individual cell nuclei was determined by absorption measurements in a rapid scanning microspectrophotometer (Lomakka, 1965) at the wavelength 546 nm. For the microscopic examination of the tumours routine histopathological techniques were employed.

RESULT

Fig. 1 shows the morphological pictures of a monomorphic adenoma and a highly differentiated

mucoepidermoid tumour of the palate. The left part of the figure shows the haematoxylin-eosin stained sections of the tumours and the right part the haematoxylin-eosin stained imprint preparations from the corresponding tumours.

Microspectrophotometric measurements of randomly selected Feulgen-stained nuclei of the imprint preparations are illustrated in Fig. 2. As control cells, human lymphocytes from peripheral blood were used. It is evident from the figure that the monomorphic adenoma cells contained the same amount of the nuclear DNA as the control cells (12–15 relative units). The mucoepidermoid tumour cells, on the other hand, show a large inter-

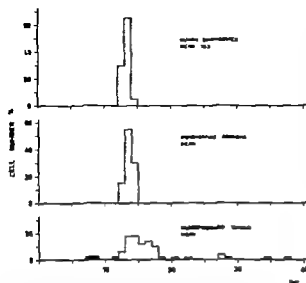


Fig 2 Intercellular distribution of nuclear DNA quantity (relative units, Feulgen-stained nuclei) in 75 human lymphocytes, 75 monomorphic adenoma cells and 75 highly differentiated mucoepidermoid tumour cells.

variation in nuclear DNA content from 7 to 38 relative units. It is important to point out that more than 50% of the analysed cells showed DNA values exceeding that of the monomorphic adenoma cells and control cells.

For purposes of comparison with a clearly malignant salivary gland tumour a DNA

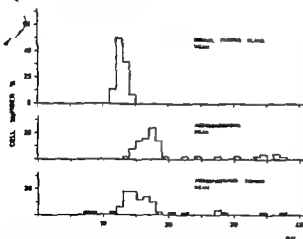


Fig 3 Intercellular distribution of nuclear DNA quantity (relative units, Feulgen-stained nuclei) in 75 normal parotid gland cells, 75 low-differentiated adenocarcinoma cells of the parotid gland and 75 highly differentiated mucoepidermoid tumour cells of the palate.

analysis was made of a low-differentiated adenocarcinoma of the parotid gland (Fig. 3). Both the mucoepidermoid tumour cells of the palate and the adenocarcinoma cells of the parotid gland show a similar increase and intercellular variability of nuclear DNA content, and both types of tumour cell populations differ greatly with respect to these DNA characteristics from the values of the normal parotid cell population.

DISCUSSION

The cytophotometric DNA analysis in the present study shows that the nuclei of the monomorphic adenoma cells had DNA values identical with those of normal control cells. The highly differentiated mucoepidermoid tumour cells, on the other hand, exhibited higher DNA values, which were comparable to those of clearly malignant salivary gland tumour cells. Thus the DNA analyses of the present study indicate that the monomorphic adenomas, despite their high cellularity are benign tumours, while mucoepidermoid tumours with highly differentiated structures should be considered as malignant and therefore be denoted as carcinomas.

REFERENCES

- Eneroth, C. M. 1971 Salivary gland tumors in the parotid gland, submandibular gland and the palate region. *Cancer* 27 1415
- Eneroth, C. M., Hjertman, L. & Moberger G. 1972a Salivary gland adenomas of the palate. *Acta Otolaryng* (Stockh.) 73 305
- Eneroth, C. M., Hjertman, L., Moberger G. & Söderberg, G. 1972b Mucoepidermoid carcinomas of the salivary glands. *Acta Otolaryng* (Stockh.) 73 68
- Eneroth, C. M. & Zetterberg, A. Microspectrophotometric DNA analysis of malignant salivary gland tumours. To be published.
- Lomakka, G. 1965 A rapid scanning and integrative cytophotometer. *Acta Histochem*, Suppl. 6, 47

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SQUAMOUS CELL CARCINOMA OF THE GINGIVA

Histological Classification and Grading of Malignancy

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Abstract. The degree of histological activity in squamous cell carcinomas of the gingiva has generally been categorized by classifying the tumours as highly moderately or poorly differentiated. This classification has in the past been generally applied despite the rather heterogeneous morphology of these types of carcinoma. Thus, the tumours are frequently highly differentiated at the surface and less differentiated in the invasive, deeper portions. The correlation between the degree of tumour differentiation and the prognosis is of limited significance and this type of classification has proved inadequate as a basis for the choice of therapy in many cases. Consequently we have deemed it urgent to try to establish a more systematic histological classification for gingival carcinomas which better reflects the biological growth capacities of the tumours and might serve as an improved aid for therapy in individual cases.

At the Department of Tumour Pathology an attempt has recently been made to classify squamous cell carcinomas of the larynx according to new principles (Jakobsson et al. 1972). Such a modified classification has been applied by us on a 12 year material of squamous cell carcinomas of the gingiva, treated at the Department of Otolaryngology and Radiumherumet, Karolinska Sjukhuset, during 1958-1969 (Nathanson et al., 1972). Altogether 125 cases have been investigated which comprise about one-third of all cases reported in Sweden during the same period.

The morphological analysis is based upon an evaluation of the tumour cell population in terms of a 1-4 point scale grading of the

degree of differentiation the nuclear polymorphism, and the frequency of mitotic figures. Separately a corresponding grading is registered of the tumour-host relationship as measured by the mode and stage of invasion as well as the degree of lympho-plasmocytic infiltration, considered as a sign of the local immunological reaction. The 6 different morphological parameters can fairly easily be estimated in routine biopsies and require only a few histological sections. The morphological analysis, though aiming to be objective, involves certain subjective factors. The reproducibility of a single investigator has, however been high. All histological examinations have been performed without any knowledge of the clinical stage of the tumour the therapy or the subsequent clinical course of the disease.

The distribution of the individual cases according to the histological grade of malignancy expressed in terms of total points values, is demonstrated in Fig. 1. There is a tendency to a bimodal distribution with a group of low-point tumours to the left and another with high-point tumours to the right. It is also obvious that patients with metastasizing tumours fall exclusively within the high-point range. There is also an intermediate group of tumours with or without metastasis. Almost all patients with metastasizing tumours had their metastasis even on admission to

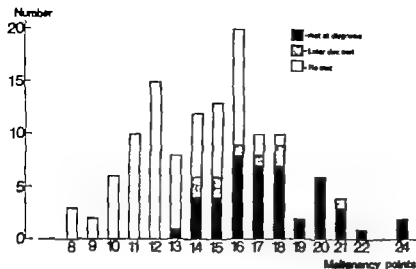


Fig 1 Histological grade of malignancy in 125 cases of squamous cell carcinoma of the gingiva, in relation to metastasis.

hospital (84%). Only a few developed metastasis later in the course of the disease (16%). This observation may suggest the presence of 2 principally different types of carcinoma, one fairly benign, which gives rise to metastasis either late or never and an aggressive, metastasizing form, analogous with what has been observed for cancer of the cervix uteri (Ashley 1966). No difference between the two groups of tumours was found in the average age of patients at onset of the disease. The distribution of metastasis in relation to the number of patients in each group and the present morphological parameters, showed, as might be expected, more frequent metastasis from tumours with high-point values. The immunological response was mostly correlated to the type of tumour cell population whereas the mode of invasion specifically paralleled the tendency to metastasis.

Dedifferentiation of the tumours during the course of the disease was rarely found in the material. Patients with multiple carcinomas in the oral cavity generally had a "finger print" response to the carcinogenic influence, reflected in a marked morphological resemblance between the different tumours.

The histological grading of malignancy presented thus implies a rather good correlation of the tendency of the tumours to metastasize. It would thus seem possible to forecast squamous cell carcinoma of the gingiva from tumour biopsies and thus make possible the institution of adequate therapy at an early stage of the disease.

REFERENCES

- Ashley D J B 1966. Evidence for the existence of two forms of cervical carcinoma. *J Obstet Gynaec Brit Emp* 73 382.
- Jakobson, P. A., Killander D, Moberger G. & Mårtensson, B. 1972. Histologisk klassifikation och malignitetsgradering vid larynx-cancer. *Föreläsningar för Svensk Förening för Medicinsk Radiologi*. In press.
- Jakobson, P. A., Eneroth, C.-M., Killander D, Moberger G. & Mårtensson, B. 1972. Histological classification and grading of malignancy in cancer of the larynx (a pilot study). *Acta Radiol* (in press).
- Nathanson, A., Jakobson, P. A. & Wersäll, J. 1972. XVIII Nordiska Kongressen i Otolaryngologi June, Göteborg.
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PROGNOSIS OF SQUAMOUS-CELL CARCINOMA OF THE GUMS

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Abstract The prognosis has been studied of 125 patients with squamous-cell carcinoma of the gums who were treated during the period 1958-1969 at the Karolinska Hospital. The material was classified according to the TNM-system (Geneva, 1968) and divided into 3 well-defined clinical stages. The determinate 5-year-survival rate was 73% in stage I (T_1-T_2), 41% in stage II (T_3-T_4) and 17% in stage III (patients with clinical metastases).

Carcinoma of the gums is a rare disease. Between 1959 and 1968, 358 new cases were registered in Sweden (Cancer Incidence in Sweden, 1959-1968) i.e., about 7% of all carcinomas of the mouth. Nevertheless, carcinoma of the gums is associated with many therapeutic problems, and opinions vary widely regarding treatment. The determinate 5-year-survival rate varies, according to the literature, between 19% (Mattick & Meehan, 1951) and 52% (Erich & Kragh, 1959). In patients with microscopically verified lymph-node metastases this figure has declined to 5% (Nathanson et al., 1971).

The aim of the present investigation was to study the prognosis of squamous-cell carcinoma of the gums in a material treated at the Department of Otolaryngology and Radiumhemmet, Karolinska Hospital, Stockholm.

MATERIAL AND METHOD

During the period 1958-1969 125 patients with histologically verified squamous-cell carcinoma of the gums were treated. Of these patients 81

were men and 44 women. In 96% of all the patients cancer occurred after the age of 50.

The primary tumour was situated in the lower gum in 82 patients and in the upper gum in 43. The primary tumour was classified according to the TNM-system adopted by the International Union Against Cancer (UICC, Geneva 1968). The material was divided into 3 clinical stages. Stage I consisted of T_1-T_2 , 22 patients. Stage II consisted of T_3-T_4 , 54 patients, and stage III comprised all the patients with palpable regional lymph-nodes, regarded as containing metastatic growth ($N+$), 49 patients.

The observation period was 1 year for 125 patients, 3 years for 123, 5 years for 110, and 10 years for 61.

The primary treatment consisted of surgery in 20 patients, radiotherapy in 73 patients (12 patients received only palliative treatment) and 32 patients received radiotherapy and were also operated but combined treatment had been planned for only 14 of these patients. In 14 patients surgery consisted in a local excision. In 26 patients radical neck dissection was performed, and in 8 patients only the sub-mandibular glands were extirpated. Hemimandibulectomy was performed in 21 patients.

RESULT

The results of the primary treatment are shown in Fig. 1 where the material is divided accord-

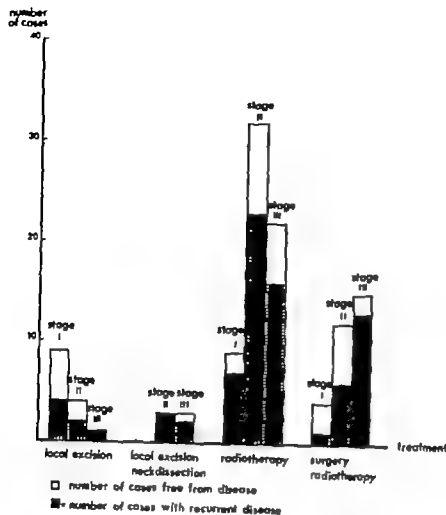


Fig 1 Results of primary treatment.

ing to the therapy given at the different stages. Here, all patients are excluded who were given palliative treatment. The best results were obtained with a local excision in stage I where 8 of 9 treated patients were still alive after 5 years, though 4 of these had a local recurrence, which was treated successfully by a new local excision. In stage II the determinate 5-year survival rate was 50% when surgery and radiotherapy were applied. In stage III the results were poor whatever form of therapy was given.

The determinate survival rate was calculated from the first histological verification of the tumour. The comparative figures for the whole material, and the clinical stages are shown in Fig. 2. For all carcinomas the 5-year-deter-

minate survival rate was 36% compared with 17% in stage III.

DISCUSSION

The results obtained in the present material support the findings in earlier materials (Martin, 1941) namely that the best results in the treatment of gingival carcinoma stage I are achieved with surgery alone. In one of 22 patients at this stage secondary metastases developed in our material. In stage II surgery and radiotherapy gave better results than surgery alone or radiotherapy alone. This is in good agreement with the recently developed concept that a combined treatment is to be preferred in cases of advanced carcinoma of the oral

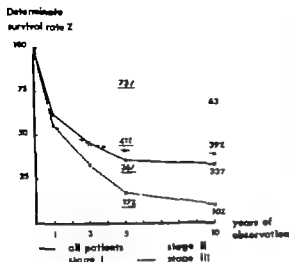


Fig 2 Survival of squamous-cell carcinoma of the gums treated 1958-1969 at the Karolinska Hospital.

cavity (Powers & Ogura, 1965). The poor results of surgery and radiotherapy in stage III is partly explained by the fact that many of these cases were not initially planned for combined therapy but were cases where radiotherapy had primarily given poor results, and surgery had then been applied, or inadequate surgery had been followed by postoperative radiotherapy. Only 6 of the patients in stage III had tumours less than 2 cm in diameter. The majority of the patients in stage III had extensive tumours with metastases, and were

regarded as a poor prognostic group also in other materials (Cady & Catlin, 1969).

It seems probable that the metastasizing tumours, with a comparatively early spread of tumour to the regional lymph-nodes, belong to an especially aggressive biological group and it is hoped that histological classification will give further information on these tumours where prognosis is especially poor.

REFERENCES

- Cady B. & Catlin, D. 1969. Epidermoid carcinoma of the gums. A 20-year survey. *Cancer* 3: 551.
- Erich, J. B. & Kragh, L. V. 1959. Results of treatment of squamous-cell carcinoma arising in the mandibular gingiva. *Arch Surg (Chic.)* 79: 100.
- Martin, H. E. 1941. Cancer of the gums (gingivae). *Amer J Surg* 54: 765.
- Metnick, W. L. & Meehan, D. J. 1951. Carcinoma of the gum. *Surgery* 29: 249.
- Nathanson, A., Bäckström, A., Jakobsson, P. A. & Wernhill, J. 1971. Prognos vid gingivalecancer med mikroskopiskt verifierade lymförtstmetastaser på hälsen. *Nord Med* (in press).
- National Board of Health and Welfare, The Cancer Registry. Cancer Incidence in Sweden 1959-1968.
- Powers, W. E. & Ogura, J. H. 1965. Preoperative irradiation in head and neck cancer surgery. *Arch Otolaryng (Chic.)* 81: 153.

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REPAIR OF THE ORAL CAVITY AFTER IRRADIATION
AND RESECTION FOR ORAL CARCINOMA

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Abstract Twenty patients with advanced carcinoma of the oral cavity were treated by major excisions of the oral cavity requiring procedures for reconstruction of the cavity walls. All patients had been treated with radiotherapy before operation. Operative procedures, postoperative complications, and results are discussed. The forehead flap was utilized in 10 cases, with very good results. The positive results support further work on combined radiotherapy and surgery in advanced cases of oral carcinoma, with use of reconstructive skin flaps when large defects are formed in the walls of the cavity.

Combined treatment with radiotherapy and surgery of advanced carcinoma in the oral cavity has been proved to give better results than either surgery or radiotherapy alone (Fletcher & Jesse, 1962; Leonard et al., 1968). When high preoperative irradiation doses are given, the rate of postoperative complications is considerably increased (Jakobsson et al., 1970; Bläckström et al., 1972). Introduction of well vascularized tissue at operation taken from areas located outside the irradiated field, will give better healing conditions and fewer complications postoperatively than surgical techniques using reconstructive procedures where only tissues from the irradiated area are utilized for reconstruction. Introduction of new tissue to cover large defects, especially in the floor of the mouth, will also reduce the postoperative deformity considerably.

The present paper deals with a follow-up study of 20 patients with advanced carcinoma

of the oral cavity treated by major surgery in which the surgical defect was reconstructed with skin flaps. All patients had received irradiation before operation.

Six patients had squamous cell carcinoma of the buccal mucosa, 7 of the gingiva, 3 were sublingual and 3 located in the tonsils. One patient had a mucoepidermoid carcinoma of the buccal mucosa. Two patients had tumours classified as T_3N_0 according to the UICC classification, 8 as $T_2N_0M_0$, 2 as $T_2N_1M_0$, 5 as $T_1N_0M_0$ and 3 as $T_1N_1M_0$ (Table I).

Seven were planned combined treatment cases and operated on 1 to 4 weeks after radiotherapy. Six cases were operated on after 2 to 6 months because of residual tumour and 2 cases respectively 9 and 10 months after radiotherapy. Five cases were operated on for recurrent disease 3 to 11 years after radiotherapy (Fig. 1).

The mean tumour dose was 5 400 rads, ranging from 2 000 to 9 000 rads, and administered for 14-71 days with 10-90 fractions. The irradiation was given with cobalt-60 teletherapy in 15 patients, with the short distance cobalt

Table I Classification (UICC)

No. of patients	20
T_3N_0	2
T_2N_0	8
T_2N_1	2
T_1N_0	5
T_1N_1	3

Supported by the Stockholm Cancer Society grant no. 71 72.

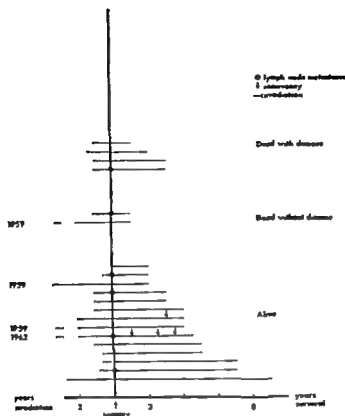


Fig 1 Survival (in years) after surgery for extensive tumours of the oral cavity

60 beam technique in 2 patients, with conventional roentgen therapy in 1 patient and with a 9 MeV electron beam in 1 patient and with a 6 MV linear accelerator in 1 patient. Five patients received 2 000–4 000 rads before operation. The majority of the group, 15 patients, received 4 500–9 000 rads (Table II).

In the earliest cases, the oral cavity defect was reconstructed using various methods, including local neck flaps, forehead flaps, Ba-

kamjian flaps or acromipectoral flaps. During the last 5 years we have more consistently used forehead flaps for inner lining of the oral cavity and local rotation flaps or Bakamjian flaps for replacement of large skin defects. Thus 10 patients had an oral lining with a forehead flap (McGregor 1963 Hoopes & Edgerton 1966), 5 patients with an acromipectoral flap, and 1 patient with a Bakamjian flap (Bakamjian & Cramer 1960). In 4 cases

Table II

	Teletherapy (60 _{Co} , kilocurie)	Short distance techniques (60 _{Co} , decacurie)	Linear accelerator (6 MV)	Electron beam (9 MeV)	Conventional roentgen
No. of patients	11	2	1	1	1
Mean dose	4 200 rad	6 600 rad	3 200 rad	5 400 rad	4 200 rad
Dose interval	(2 000–9 000) rad	(6 500–6 700) rad	—	—	—
Treatment	15–71	28–34	33	18	26
No. of fractions	10–90	24–25	22	12	23

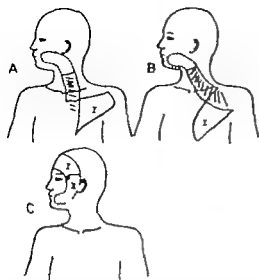


Fig. 2 Schematic figure illustrating the rotation of flaps for covering of defects in the oral cavity. The Bakamjian flap and the acromioclavicular flaps form a fistula on the neck which is closed secondarily. The forehead flap is folded under the skin of the zygoma area and forms a fistula upward which is closed when the part of the flap not used for the covering of the defect in the oral cavity is put back on the forehead area. A. Bakamjian flap. B. Acromioclavicular flap. C. Forehead flap.

rotation of a local flap from the neck was sufficient for inner lining (Figs. 2, 3 Table III).

Of 20 operated cases 12 had postoperative complications with fistula formation (9) paraspinal flap necrosis (3) bone necrosis (1) or other necrosis (3). All complications healed either spontaneously or after further surgery and did not cause major distress for the patient (Table IV).

The patients were followed from 2 to 12 years postoperatively. During the follow-up period 4 patients died of cancer, 2 patients died without signs of cancer and 14 patients are alive more than 2 years after surgery with a mean survival time of 4.5 years. All patients

Table III. Type of reconstruction

Neck flap	4
Forehead flap	10
Acromioclavicular flap	5
Bakamjian flap	1

Table IV. Complications

No. of patients	12
Fistula	9
Partial flap necrosis	3
Bone necrosis	1
Skin necrosis	3

who died of cancer died within the first 4 years after surgery (Table V).

The results of treatment in this series of patients with advanced carcinoma of the oral cavity has been encouraging. We have demonstrated that major excisions of the oral cavity can be performed, and large defects reconstructed with skin flaps after radical doses of preoperative irradiation, without severe complications in the postoperative course. The use of forehead flaps or combinations of forehead flaps with other flaps decreases the fibrosis in the tissues of the oral cavity and the postoperative problems with swallowing and eating. Introduction of well vascularized tissue from areas outside the irradiated field reduces complications after surgery does not increase operative risk, even in the elderly patient.

REFERENCES

- Bakamjian, V. & Cramer, L. 1960. Surgical management of advanced cancer of the tongue. *Ann Surg* 152 1058.
- Bäckström, A., Jakobsson, P. A., Ljundberg, B., Sandquist, S. & Wernvall, J. 1972. Postoperative complications after combined treatment for carcinoma of the tongue. *Arch Klin Exp Ohr Nas Kehlkopfheilk* 201 273.
- Fletcher, G. H. & Jense, R. H. 1962. The contribution of supervoltage roentgenotherapy to the integration of radiation and surgery in head and neck squamous carcinomas. *Cancer* 15 566.

Table V. Results

No. of patients	20
Died of cancer	4
Dead of intercurrent disease	2
Alive	14
Mean survival time years	4.5

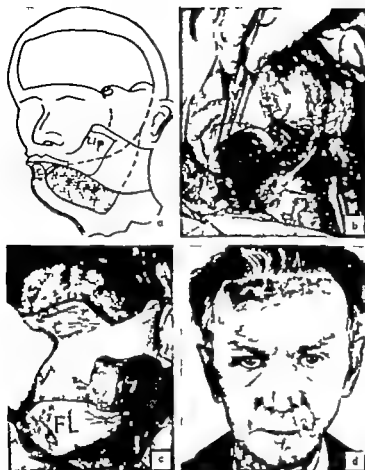


Fig 3 The forehead flap is wide enough to cover large parts of the floor of the mouth. A illustrates schematically the procedure used in covering of the large defect in the floor of the mouth formed when a large gingival carcinoma T was operated with resection of the mandible from the area of dens to the angle of the mandible of the left side with left

side of the tongue and floor of the mouth. A radical neck dissection was made at the same time. B shows the defect. The tongue is held by forceps. C shows the forehead flap (FL) in place before suturing down the flap including the lower lip which was cut in the midline (cf. A). D shows the patient 2 years after operation.

Hoopes, E. & Edgerton, M. T. 1966 Immediate forehead flap repair in resection for oropharyngeal cancer. *Amer J Surg* 112 527.

Jakobsson, P. A., Litbrand, B., Sandquist, S. & Westin, J. 1970. Kirurgiska komplikationer efter preoperativ strålbehandling av tungcancer. *Nord Med* 83 647.

Leonard, J. R., Litton, B. W., Latourette, H. B. & McCabe, B. F. 1968. Combined radiation and surgical therapy: tongue, tonsil and floor of the mouth. *A. Otol* 77 514.

McGregor, I. A. 1963. The temporal flap in intraoral cancer: its role in repairing the post-resectional defect. *Brit J Plast Surg* 16 318.

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TOLUIDINE BLUE STAINING AS A MALIGNANCY TEST

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Abstract In order to get a reliable material at the cytological sampling or at the biopsy the Toluidine blue test has been designed. The method is based upon the fact that cancer cells contain quantitatively more DNA and RNA to which nucleic acids the Toluidine blue has an affinity. For that reason an oral staining with that metachromatic dye delineates the region of malignancy of the epithelial mucosa. A consecutive study of 54 stained lesions was reported. The results were correlated to the histopathological investigations and proved to agree.

The reliability of the cytologic and the histopathologic diagnosis of cancer depends on the representativity of the tissue sent to the pathologic service.

The Toluidine blue test has been designed as a method to delineate the region of malignancy in a suspected lesion, which could hide areas essential to the pathologic investigation.

The method is based upon the fact that cancer cells contain quantitatively more RNA and DNA. Toluidine blue has an affinity to these nucleic acids, and for that reason the malignant tissue will stain more strongly than will the normal epithelial tissue. At the Department of Oral and Jaw Diseases, Karolinska Hospital, this test has been utilized during the last 4 years. In this technique, factors which could weaken the reliability of the test were avoided.

The stain was used in a 1% water solution, the temperature of which was kept within a range of 15–20 degrees, and the pH value of the lesion was about 5.

The lesions were cleaned and dehydrated in 1% acetic acid. The stain was applied abundantly to the lesion in question for 30 seconds. Decolourization was performed with 1% acetic acid. The staining results were then determined. If the lesions retained a deep blue or violet colour tone the test was considered positive.

A consecutive study of 54 lesions was performed. The results of the test were correlated to the histopathologic investigations of the same lesions and the results proved to agree. There was a small difference only in the dysplasia group. The staining of the cancer cells was also studied by the technique of nitrogen-frozen sections (in collaboration with Dr Roger Willén, Department of Tumour Pathology Radiumhemmet, Stockholm). In this study the staining of malignant nuclear structures was remarkable.

REFERENCES

- Shedd, H. P. et al. 1967. Further appraisal of *in vivo* staining properties of oral cancer. *Arch Surg (Chic.)* 93: 16.
Strong, M. E. et al. 1968. Toluidine blue in the management of carcinoma of the oral cavity. *Arch Otolaryng (Chic.)* 87: 101.

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TONGUE CANCER

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The Cancer Registries of Finland, Iceland, Norway and Sweden have recently published a report on cancer incidence in these countries. According to these statistics Finland shows a decided male preponderance in total cancer incidence otherwise the differences between the four countries are small. The incidence of stomach cancer is very high in Iceland and also higher in Finland than in Norway and Sweden. On the other hand, the latter two countries have higher incidence of cancer of colon and rectum than Finland and Iceland. Sweden shows the highest rates in cancer of liver and biliary passages. The most striking feature in cancer of respiratory organs is the high male lung cancer incidence in Finland (more than 5-fold as compared with that in Iceland). On the other hand, in females Iceland has the highest rate, and the Finnish rate is equal to that of Sweden.

In laryngeal cancer too the Finnish male rate is high (3-4-fold as compared with that of the other countries). The Finnish rate for female breast cancer is only about two-thirds of the rates for the other countries. Carcinoma of skin is high in Finland. Iceland has a high rate of thyroid cancer.

Table I shows the relative incidence of tongue cancer in Finland, Iceland, Norway and Sweden. The incidence rates are counted per 100 000 of population in Iceland 1957-66, in Finland, Norway and Sweden 1962-66.

The total incidence of tongue cancer is 50% higher in Finland than in Sweden. The rates for Iceland and Norway lie in between these two. In Iceland the rate for women is higher than for men. In the other countries the order is the reverse.

The high incidence of tongue cancer in Finland may be related to dental caries and deficient hygiene of the mouth. Smoking habits are probably also an important factor.

Table I. Incidence of tongue cancer

Incidence	Finland	Iceland	Norway	Sweden
Male	0.9	0.5	0.9	0.6
Female	0.6	0.8	0.4	0.4
Total	1.5	1.3	1.3	1.0

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CARCINOMA OF THE LIP

A Series of 869 Patients

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Abstract In a series of 869 patients with carcinoma of the lip the principles of treatment of the primary tumours and the regional lymph node metastases are discussed. The results of treatment are presented.

Between 1946 and 1966 a series of 869 patients with carcinoma of the lip were seen in the Radium Center and the E.N.T. Department in Aarhus.

The present report deals with the problems and the results of treatment of both the primary tumour and the metastases. 97.2% of the patients were males. As regards the age distribution the incidence was greatest in the group aged 60-69. All cases were squamous-cell carcinomas, 8 of these were basocellular carcinomas. A rather large number of the patients (57%) had the primary lesion for more than 4 months. In only 1.8% (16/869) of the patients was the tumour located on the upper lip. The series was grouped according to the TNM classification in the UICC modification. There was a great preponderance of early cases, only 111 patients being T3 and T4 cases. In 14 patients regional lymph node metastases were found on admission. None of the patients had distant metastases.

The principles of treatment of the primary tumour can be seen in Table I.

165 recurrences, residual tumours and new tumours occurred in 132 patients. In only 7 patients was it impossible to control the primary local tumour. In a little more than half

of the patients, secondary treatment was radiotherapy; the rest underwent various operations.

The Radium-needle intubation failed in only 11% of the cases, but if the intubated T3 cases are regarded selectively the recurrence rate was found to be 25%. Therefore, the T3 cases ought not to be treated by intubation. Because it is possible to control the primary tumour in the majority of the patients the malignant character of the carcinoma of the lips is determined by the tendency to metastases.

The incidence of the regional lymph node metastases in this series can be seen in Table II.

Most of the metastases were located in the submental or submandibular lymph nodes and no metastases jumped those primary lymph node stations.

TREATMENT

The results of treatment can be seen in Table III.

The two groups in Table III are not comparable. In the irradiation-group relatively

Table I

Intubation (10 mg radium needles)	766
External radiation	74
Operation	29
Total	869

Table II

Total no. of cases	Nodes present on admission	Node involvement at subsequent follow-up	Total node involvement at any time	No node involvement at any time
869	14 (1.6%)	45 (5.2%)	59 (6.8%)	810

Table III

	No of cases	Dead from cancer laibl	Dead from intercurrent disease	Crude 5-year survival rates
Neck dissection	37	9	3	68% (25/37)
Irradiation	22	16	1	23% (5/22) ^a
Total	59	25	4	51% (30/59)

^a One patient alive at 5-year limit with recurrence.

more of the metastases were fixed than in the group of surgical treatment. The cases with histologically verified metastases in the surgical group had a 5-year crude survival rate of 56% (15/27). Fifteen patients in the surgical group had only partial neck dissections, and recurrences were seen in 9 of those patients. In 4 of these 9 patients the regional metastases were controlled by total neck dissections. There is reason to believe that total neck dissection

as the initial attempt at treatment might have reduced the recurrence rate.

Prophylactic neck dissection was not performed. However an analysis of the T3-cases showed that T3-cases with recurrence of the primary tumour of the lip had a frequency of lymph node metastases of 38%. Therefore a prophylactic exploration of the submental and the submandibular lymph nodes should be considered in this special group. If freezing microscopy proves malignancy in the nodes a total radical neck dissection should be performed.

Five-year crude survival rate of the whole series was $84.5 \pm 2.5\%$. After correction for mortality from other causes than cancer the 5-year survival rate was found to be 96.7% i.e. only between 3 and 4% of the patients die from carcinoma of the lip.

In Fig. 1 the survival corrected curves for the T1, T2 and T3 cases are shown.

Thus, most cancer deaths are found in the T3 group, as mentioned, not because of the primary tumour in itself but because of a greater frequency of lymph node metastases.

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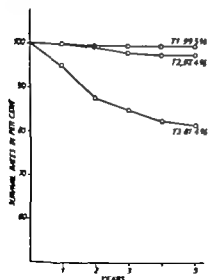


Fig. 1

CARCINOMA OF THE LIP

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treatment. The treatment established against the recurrences was dependent on the initial therapy and could be surgical or radiotherapeutical alone or in combination. A very few patients received chemotherapy or no therapy at all.

The 5-year survival rate of the total material was 45% (43 patients) with the following distribution in the different groups according to the TNM system.

T1	T2	T3	T4
14 (64%)	11 (46%)	10 (42%)	8 (32%)

No difference was found between males and females. Furthermore, 7 patients died of intercurrent diseases within 5 years without signs of malignant tumour.

In spite of the small material analysed here we wish to stress a few factors which might be of importance in raising the percentage of 5-year survivals.

1 All patients with cancer of the tongue irrespective of its grade should be treated sur-

gically. A combined therapy surgical plus radiotherapeutical, could also be performed. The size of the surgical intervention should at least be partial glossectomy and a homolateral neck and submandibular dissection. In most cases a contralateral submandibular dissection must be considered, especially when the tumour is located near the midline of the tongue.

2. In patients with tumours above T2 and in all patients with palpable nodes in the submandibular region a partial mandibular resection should also be done.

3. Where resection of the mandible has been performed, a space-occupying construction or transplant ought to be discussed. We think this is an important procedure in the postoperative rehabilitation of the patient.

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STROMAL REACTIONS AS PROGNOSTIC FACTORS IN EPIDERMOID CARCINOMA OF THE TONGUE

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Abstract The appearance and clinical significance of round cells and acid mucosubstances in the tumour stroma were investigated in 80 patients with epidermoid carcinoma of the tongue. It was found that on the average the prognosis was better in the cases where the round cell reaction in the tumour stroma was positive, and also in the cases where the acid mucosubstance reaction was negative in the tumour stroma. Furthermore, an inverse correlation between the appearance of round cells and acid mucosubstances was noted.

A malignant epithelial neoplasm forms a functional whole with the surrounding mesenchymal tissue. Obviously the clinical characteristics of the carcinoma depend on both the parenchyma and the stroma. In the earlier literature opinions differed as to the significance of the round cell reaction of the stroma for the prognosis of the patient. Most recent authors have found that a strong round cell reaction is often connected with a better prognosis (Hayashi et al., 1964).

Differing amounts of acid mucosubstances have been found in the stroma of malignant neoplasms. According to a recent investigation there is an inverse correlation between the appearance of mucosubstances in the stroma and the prognosis of the patient (Paavolaime, 1970).

No mucosubstance investigations have been made on neoplasms in the oral cavity. In the present investigation the appearance as well as the significance of both acid mucosubstances and round cells have been investigated in paral-

lel in a material comprising 80 epidermoid carcinomas of the tongue.

MATERIAL AND METHOD

The material comprises 35 men and 45 women suffering from epidermoid carcinoma of the tongue. The mean age of the patients was 69.5 years. Some of the patients were subjected to radiotherapy alone, some to surgery alone, and some of the patients to both modes of treatment. The follow-up period was 5 years. The prognosis was estimated on the basis of the survival of the patients.

The round cell reaction was studied using the Weigert van Gieson staining method. The mucosubstances were stained with Hale's Colloidal Iron method.

RESULT

The following table (Table I) presents the correlation between round cell reaction and prognosis.

It is evident from the table that on the average patients with positive round cell reaction had a better prognosis.

Table II shows the correlation between acid mucosubstances and prognosis.

It can be seen that on the average the prognosis was better with patients with negative mucosubstance reaction.

Table I. Survival by occurrence of round cell reaction

Round cell reaction	No. of cases	Survival	
		No. of cases	%
Absent	32	8	25
Present	48	20	41
Total	80	28	

The following table (Table III) presents the correlation between appearance of both round cells and acid mucosubstances.

It is evident from this table that in the group where there was no mucosubstance reaction, the round cell reaction was clearly more often positive as compared with the group with positive mucosubstance reaction. In other words, acid mucosubstances and round cells seem to avoid each other's vicinity.

DISCUSSION

The number of patients is so small that statistical conclusions are difficult to make. However these results agree with those obtained in a large laryngeal carcinoma material at our clinic (Paavola, 1970).

One possible interpretation of the results of the present investigation is the following: the mucosubstance coat surrounding the carcinoma acts as a barrier which prevents the cell-bound immunity influence on the tumour. Thus carcinomas which lack this barrier can more easily be destroyed by the organism itself. The theory presented by Currie & Bagshawe (1967) and

Table II. Survival by occurrence of acid mucosubstances

Mucosubstances	No. of cases	Survival	
		No. of cases	%
Absent	25	11	44
Present	55	17	31
Total	80	28	

Table III. Number and percentage distribution of cases by round cell reaction and occurrence of acid mucosubstances

Round cell reaction	Occurrence of mucosubstances			
	Absent		Present	
	No. of cases	%	No. of cases	%
Absent	5	20	27	49
Present	20	80	28	51
Total	25	100	55	100

Kirby & Wood (1967) gives an interesting background for the interpretation of the results. It has been found that acid mucosubstances appear except around carcinomas also on the surface of the trophoblasts of the placenta. Carcinomas and embryos are the only "foreign bodies" that the organism cannot effectively reject. As is known, carcinoma and embryo cells are morphologically similar to each other. On the other hand, some specific antigens are known to occur both in embryonic tissue and in carcinoma cells. It might be that the organism is not able to distinguish between a carcinoma and an embryo and thus begins to protect the carcinoma as the embryo by means of mucosubstances against its own mechanisms of rejection.

REFERENCES

- Currie, G. A. & Bagshawe, K. D. 1967. The masking of antigen on trophoblast and cancer cells. *Lancet* **i** 708, 1110, 1272, **ii** 1336.
- Hayashi, S., Kawamata, K., Tachikawa, I., Takada, Y., Watanabe, T., Iwama, N. & Tsuda, H. 1964. Stromal reaction and prognosis in cancer of the stomach. *Geka Shiryō* **6** 835.
- Kirby, D. R. S. & Wood, C. 1967. Embryos and antigens. *Science* **3** 56.
- Paavola, M. 1970. Stromal reactions as prognostic factors in epidermoid carcinoma of the larynx. M.D. Thesis (Helsinki).

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TREATMENT OF SQUAMOUS CELL CARCINOMAS IN HEAD AND NECK WITH BLEOMYCIN AND WITH COMBINED BLEOMYCIN AND X-RAYS

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Abstract. The choice of therapy for patients with squamous cell carcinoma will depend of the histology of the tumour its extent and localization. X-rays and surgery alone and combined, are well established methods by which elimination of the tumour may be achieved. With a new therapeutical approach, combining bleomycin and X-rays, a possible synergistic effect has been obtained. It has in itself given surprisingly good results and when used as a pre-operative treatment it offers possibilities of making an increased number of otherwise inoperable cases surgically accessible.

Bleomycin has been shown to have a significant therapeutic effect on squamous cell carcinomas, particularly those of the highly differentiated type. In contrast to other cytostatics it does not affect the bone marrow nor has it an immunosuppressive effect.

At the Department of Otolaryngology of Rikshospitalet, bleomycin has been used in about 100 cases of squamous cell carcinoma of the head and neck. To begin with we administered the usual dosage pattern of 30 mg i.v. twice per week for 5-6 weeks. Based on animal experiments (Juul Jørgensen, 1971) and our own clinical observations we changed, 6 months ago to a combination treatment, bleomycin and X-ray.

Before going any deeper into this new approach, we should report our experience with respect to the therapeutical efficacy and side-effects with bleomycin alone (Tables I, II).

One of the remarkable cases is a recurrent laryngeal carcinoma after radiotherapy. Following the bleomycin treatment this tumour disappeared completely now 2½ years ago. We would also like to mention 8 cases of carcinoma of the oral cavity and tongue which showed a profound regression. A tumour regression >50% often made it possible to remove the remainder by surgery. In our experience bleomycin's most important field is pre-surgical chemotherapy.

The surgery should be performed shortly (2-3 weeks) after the bleomycin treatment is completed.

The side-effect to cause most concern has been an interstitial pneumonia. We had only 2 cases, both with moderate symptoms. We cannot account for this apparent difference between our and others experience. A point of possible importance is to keep a slow rate of i.v. injection.

We have seen no indications of damage to the inner ear.

Experiments in mice with i.m. inoculated methylcholanthrene induced epithelial carcinomas. Juul Jørgensen (1971) has shown that simultaneous combination of bleomycin and X-rays has significantly increased the therapeutical efficacy of the treatment. On the background of this study and our own experience with bleomycin as well, we have

Table I. Response of tumour to bleomycin treatment in patients with squamous cell carcinoma

Localization	No. of patients	Regression of tumour			
		Complete	>50%	<50%	None
Larynx	15	1		13	1
Oral cavity	8		4	3	1
Tongue	8		4	4	
Hypopharynx/oesophagus	5		1	4	
Epipharynx	1		1		
Nose/Sinuses	3		2	1	
Ear/Temporal bone	4		2	2	
Lacrimal sac/Orbit	1		1		
Skin (ext. ear)	2	1	1		
Long metastases	1				1
Total	48	2	16	27	3

Table III. Squamous cell carcinomas response of tumours to combined treatment with bleomycin and X-rays

Localization	No. of patients	Regression of tumour			
		Complete	>50%	<50%	None
Larynx	17	11	5	1	
Oral cavity	2	2			
Tongue	2	1		1	
Hypopharynx/oesophagus	5	1	3	1	
Epipharynx	2	1	1		
Nose/Sinuses	5	2	2	1	
Ear/Temporal bone	2		1	1	
Parotid gland	1	1			
Skin (ext. ear)	1	1			
Inoperable neck metastases (laryngeal 7 cases) (tonsillar 2 cases)	9	6	3		
Total	46	26	15	5	

since November 1971 combined bleomycin with X-ray treatment.

Our schedule was as follows.

First week. Bleomycin i.m. 15 mg daily Mon.-Sat., given 1 hour before X-rays. X-rays, 350 R daily Mon.-Sat.

Second week. Bleomycin i.m. 15 mg Mon., Wed., Fri.

Third week. Bleomycin i.m. 15 mg daily Mon., Wed., Fri. given 1 hour before X-rays. X-rays, 350 R daily Mon.-Sat.

Total dosage: Bleomycin 180 mg. X-rays, 4 200 R.

The therapeutical response is shown in Table III.

Carcinoma of the oral cavity and tongue constitutes a relatively small group, but the results are very good. In the group of inoperable neck metastases complete regression was obtained in 6 cases and more than 50% regression in 3 cases. A total of 26 complete

regressions out of 46 cases is also remarkable. Whether the effect of the combination treatment is synergistic or cumulative is difficult to decide. In patients with previously untreated tumour we have observed a conspicuously rapid regression. This fact and the results in patients with inoperable metastases in the neck, strongly indicate synergism. We must emphasize, however that these are only early results.

The side-effects we observed with the combination treatment are essentially the same as the typical side-effects for bleomycin alone and X-rays alone.

During the study of biopsies from bleo-

Table II. Treatment with bleomycin side effects

No. of patients	Swelling of skin	Stomatitis	Hair loss	Fever	Pruritus Erythema Pigmentation	Pulmonary lesions
56	38	29	20	7	6	1

Treatment stopped owing to skin lesions (necrodermis): 5 cases.

Treatment stopped owing to stomatitis: 1 case.

mycin-treated patients, we have observed changes that may be of great significance. Islands of tumour cells are found where undifferentiated "basal-cell-like" tumour cells are no longer visible. Such "Basal-cell-like" cells are ordinarily observed in untreated squamous cell carcinomas, and in squamous cell carcinomas treated with other cytostatics or with irradiation alone. In bleomycin-treated patients, where a good clinical effect is seen islands of cells are found that seem to have undergone maturation very rapidly. Some of the nuclei have not lost their stainability and histology shows streaks of keratin or keratin-like substances with pycnotic nuclei similar to what in psoriasis is called parakeratosis. Around such islands of keratin are often found multinucleated giant cells of the foreign body

type. Very few lymphocytes are seen. This may be interpreted as showing that bleomycin, in addition to its effect on cell proliferation probably also affects the keratinization process and speeds it up or changes it qualitatively. This may be the reason why bleomycin has such good effect on squamous cell carcinomas, especially the highly differentiated types.

REFERENCE

Juul-Jørgensen, S. 1971. Dose schedules and combination with radiotherapy in bleomycin treatment. Paper read at the VII Int. Congr. of Chemotherapy, Prague, August 1971.

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COMBINED RADIOTHERAPY AND SURGERY IN TREATMENT OF CARCINOMA OF THE TONGUE

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Abstract Twenty-three patients with carcinoma of the tongue were treated with combined radiotherapy and surgery. A follow-up study was made, demonstrating a 55% three-year determinate survival rate and a 43% five-year determinate survival rate. Postoperative complications were found only in patients treated preoperatively with more than 4 500 rads.

The variation in stages, location, and mode of therapy of carcinoma of the tongue makes it difficult to compare treatment results in various series. It is quite obvious, however, that there has been an improvement in treatment results during recent decades. The 5 year determinate survival rate for treatment of all cases of cancer of the tongue and floor of mouth lies around 40% whether surgery or radiotherapy is used (Harold, 1969; Montana et al., 1969). The cure rate falls rapidly from about 80% in small lesions of the anterior part of the tongue without metastasis, to less than 15% in large posterior tongue lesions with metastasis.

The aim of the combined use of radiotherapy and surgery is to reduce the tumour volume preoperatively by destroying the largest possible part of the tumour cell population and thereby increasing the chances for radical extirpation of the tumour. A decreased risk of spreading tumour cells or reimplanting the tumour during operation is also aimed at. Few controlled series with treatment results have, however, been published. A plea for collaborate studies in preoperative radiation was made by the

editor of the Journal de l'Association Canadienne des Radiologistes in 1965.

The increased risk of postoperative complications has been an obstacle which has made many surgeons unwilling to operate after irradiation. Refined techniques in radiotherapy as well as in surgery have, however, considerably reduced the risk of postoperative complications. In a recent publication Bäckström et al. (1972) demonstrated that there was no increase in postoperative complications, as compared with non-irradiated material, when the preoperative radiation dose was 4 000 rads or less, and given as a fractionated treatment over a period of 21 days.

Combined radiotherapy and surgery has been used consistently for the treatment of all cases of advanced carcinoma of the tongue treated at the Karolinska Sjukhuset during the last 5 years, and for selected cases during more than 10 years. All patients were seen by a joint group of radiotherapists and surgeons.

A follow-up study was made of 23 cases of carcinoma of the tongue treated during the years 1961 to 1970. The tumours were classified according to the UICC classification. Nine of the patients had advanced carcinomas classified as T₃ or T₄, and 4 were classified as T₁ or T₂. Six patients had cytologically verified metastases in the neck prior to treatment.

All patients received preoperative radiotherapy. The mean tumour dose was 4 400 rads (range: 3 800-6 700 rads) which was administered over 12 to 56 days, using 10 to 43 fractions. The irradiation was given with cobalt-60 teletherapy in 21 patients and with short dis-

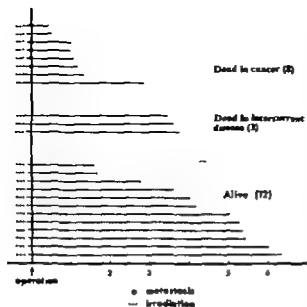


Fig 1 Survival (in years) after combined therapy for cancer of the tongue.

tance cobalt-60 beam technique in 2 patients. All patients were operated upon within 3 months of completion of radiotherapy. In 16 patients the operation consisted of partial excision of the tongue and neck dissection on one side. Seven patients had a partial excision of the tongue only.

The follow-up period varied between 2 and 11 years (mean 5.8 years).

Eight patients died of cancer during the observation period (Fig. 1). Three patients died

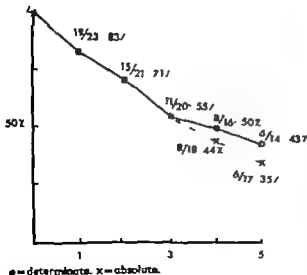


Fig 2 Survival, expressed as a percentage and in number of years after treatment.

of intercurrent disease and 12 are alive. The 3 year determinate and absolute survival rate was 55%. The 5 year determinate survival rate was 43% and the absolute survival rate was 35% (Fig. 2).

None of the 14 patients treated with 4500 rads or less had any postoperative complications. Nine of these are alive without tumour. 4 died of cancer less than 2 years after operation, and one died of intercurrent disease 4 years after treatment. Among the 9 cases that received doses higher than 4500 rads there were 2 with some necrosis of the oral mucosa, one with a cutaneous fistula, and one patient had difficulty in swallowing during the first month. The fistula healed without operation. Five of these patients survived more than 3 years without cancer. All of these patients carried on a normal life less than 4 months after operation. Two of the 5 patients died of intercurrent disease 3 1/2 years after operation.

In conclusion, we feel that a radiation dose of 4000 rads or less given over 21 days to the oral cavity does not increase the risk of postoperative complications and is well tolerated by the patient. The treatment results are promising and support further work along the same line. We are convinced that pre-operative radiotherapy is a valuable adjunct to surgery in the treatment of advanced carcinoma of the tongue.

REFERENCES

- Bäckström, A., Jakobsson, P. A., Litzbrand, B., Sandquist, S. & Weräll, J. 1972. Postoperative complications after combined treatment of carcinoma of the tongue. *Arch Klin Exp Otor Nas Kehlkopf heilk* 201: 273.
- Harold, C. C. 1969. Cancer of the tongue: some comments on surgical treatment. In *Symposium on cancer of the head and neck* (ed. John C. Gafford), p. 185. C. V Mosby Company St. Louis.
- Montana, G. S., Hellman, S., von Esen, C. F. & Kilgerman, M. M. 1969. Carcinoma of the tongue and floor of the mouth. *Cancer* 23: 1284.

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BLEOMYCIN THERAPY AND OTOTOXICITY

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Abstract Bleomycin produced regression of tumours in 10 of 15 cases of human squamous cell carcinoma. Hearing and equilibrium were not affected. No ototoxic effects were found in guinea pigs when Bleomycin was administered subcutaneously and intraperitoneally. Cochlear damage was seen when the drug was injected into the middle ear.

Fifteen cases of squamous cell carcinoma were treated with an average total dose of 300 mg of Bleomycin. Results of this treatment are shown in Table I.

Good tumour regression was observed in 3 of the cases. In one of these remission has lasted for over a year. In 5 of the cases lung complications occurred, one of them being fatal. Other side effects noted were nausea, skin and mucosal lesions, hair loss, and fever. In 2 cases acute bleeding gastric ulcers occurred during treatment.

In 6 controlled cases, vestibular function and hearing were found to be unimpaired by the treatment.

Since some antitumour agents such as nitrogen mustard in high concentration have been shown to be ototoxic (Cummings, 1968) and since Bleomycin is an antibiotic of the glucosamide group (Umezawa et al. 1966, Ishizuka et al. 1967), some members of which have serious ototoxic effects (Kohonen, 1965), an experimental investigation was conducted into the ototoxicity of Bleomycin. Three groups of guinea pigs were exposed to the drug by

different routes of administration. Group 1 and 2 received 35-40 mg subcutaneously and intraperitoneally respectively over a period of 1 week. Group 3 received 1-2 mg in 0.1 ml saline injected into the tympanic membrane. Animals in Group 1 and 2 which had received a total dose ten times greater than that used in cancer therapy showed no significant physiological ototoxic effects. Histological surface specimen technique (Edsmyr, 1966) supplemented by scanning electron microscopy. Animals in Group 3 had inner hair cell losses when examined 1 week after treatment. Histological investigation showed that the inner hair cells had shorter survival times than the outer hair cells. Larger doses of Bleomycin caused severe damage to the inner hair cells and inner hair cells. Inner hair cells did not appear to be damaged. Damage was most pronounced at the base of the cochlea and decreased toward the apex. Similar effects have been reported as a characteristic of some antineoplastic (Kohonen, 1965).

Peroxidase-benzidine staining was used to investigate the effect of Bleomycin on the stria vascularis. Group 1 and 2 animals showed no effects in the animals of Group 3 general atrophy of the stria was observed. In one of the Group

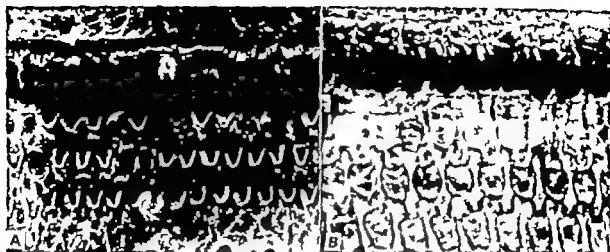


Fig. 1 Hair cell damage in the organ of Corti of a guinea pig after injection of Bleomycin into the middle ear. The scanning electron micrograph (A) reveals that all three rows of outer hair cells are

affected. The phase contrast micrograph (B) emphasizes the sensitivity of the cells in the first row (arrow).

circumscribed lesion characterized by sharp interruption of several stria capillaries and the formation of fibrous capillary strands, was found in the third coil. Further investigation is required to determine if this lesion represented an effect of the Bleomycin or had some other etiology.

CONCLUSION

Bleomycin which appears to have some value as an antitumour agent in humans, was found to have no ototoxic effect on guinea pigs when

administered in the normal parenteral way in doses per weight unit far exceeding those used in human antitumour therapy. However, hair cell losses were found in guinea pig cochleas when the drug was injected through the tympanic membrane.

REFERENCES

- Cummings, C. W. 1968. Experimental observations on the ototoxicity of nitrogen mustard. *Laryngoscope* 78: 530.
 Engström, H., Aden, H. W. & Andersson, A. 1966. *Structural pattern of the organ of Corti*. Almqvist & Wiksell, Stockholm.
 Ishizuka, M., Takayama, H., Tatenchi, T. & Umezawa, H. 1967. Activity and toxicity of Bleomycin. *J. Antibiot. (A)* (Tokyo) 20: 15.
 Kobonen, A. 1965. Effect of some ototoxic drugs upon the pattern and innervation of cochlear sensory cells in the guinea pig. *Acta Otolaryng. (Stockh.)*, Suppl. 203.
 Umezawa, H., Maeda, K., Tatenchi, T. & Okada, U. 1966. New antibiotics, bleomycin A and B. *J. Antibiot. (A)* (Tokyo) 19: 200.

Table 1 Effect of Bleomycin in 15 cases of squamous cell carcinoma

Localization	No. of patients	Regression of tumour			
		Complete	> 50	< 50	None
Tongue	5	—	1	2	2
Oral cavity	2	—	1	—	1
Nasopharynx	2	—	1	1	—
Hypopharynx	3	—	—	1	2
External auditory canal	4	—	—	2	—
Skin	1	—	—	1	—
Total	15	0	3	7	5

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AUDIOLOGICAL AND MORPHOLOGICAL ASSESSMENT OF EFFECT OF NOISE ON COCHLEA AND BRAIN STEM IN CAT

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Part 1 *Audiological Assessment* (G Lidén)

The pathogenesis of the loudness recruitment phenomenon has long been of considerable interest. There are few clinical reports on this matter based on autopsy observations. The purpose of this experimental study on cats was to correlate noise-induced hearing loss and recruitment to underlying structural changes in the cochlea and in the brain stem.

Eleven unilaterally labyrinthectomized cats were exposed to broad band noise of 115 dB SPL continuously for 8 hours. The tone thresholds before, immediately after and finally 3½ months after exposure were determined with avoidance conditioning technique.

Recruitment of loudness was considered to be present in the cat with induced hearing loss if the stapedia reflex could be elicited at the same level as before exposure. The reflex was elicited by means of sound stimulation in the same ear in which the reflex was measured, in accordance with the method given by Møller 1960.

The results showed an average hearing loss of 45 dB in the frequency range 500-2 000 Hz. The induced loss did not change the thresholds of the stapedia reflex, thus indicating the presence of loudness recruitment. The details of the audiological part of this study has been published elsewhere (Lidén 1970).

REFERENCES

- Lidén, G 1970. The stapedius muscle reflex used as an objective recruitment test. In *Ciba Foundation Symposium on Sensorineural hearing loss*, pp 295-311 Churchill, London.
- Møller Aa. 1960. Improved technique for detailed measurements of the middle ear impedance. *J Acoust Soc Amer* 32 250.

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Part 2 *Effect on Inner Ear Sensory Cells* (H. Engström)

The structural modifications resulting from noise exposure have been studied by the surface specimen technique (Engström et al., 1966) and in a few cases also by scanning electron microscopy (Engström & Engström, 1972).

RESULT

In all animals except one (LL27) we have made a practically complete preparation of

the organ of Corti and recorded the presence or absence of almost all outer and inner hair cells. In spite of the pronounced loss of hearing as recorded above the extent of hair cell loss is rather small. In the first and second row of outer hair cells the percentage of lost cells is less than 2% while the third row has a somewhat more pronounced sensory cell loss. In certain areas this cellular destruction is more pronounced but in no animal is there a

complete loss of sensory cells such as we have often found after exposure to impulse noise (Bredberg et al., 1972)

DISCUSSION

In this series of unilaterally "labyrinthectomized" cats exposure to noise has caused a considerable hearing loss, as found by audiometric examination (avoidance technique). This hearing loss is paralleled by a morphological modification of the organ of Corti with loss of sensory cells, especially in the third or outermost row of hair cells. The loss is less extensive than expected and this is in good agreement with the results of several recent observers (cf Hunter-Duvar 1973) indicating that a considerable hearing loss may be found in spite of a moderate loss of sensory cells. This indicates that functional modifications causing hearing loss may appear inside the organ of Corti while most hair cells are still

present. Studies under the scanning electron microscope indicate that small modifications, difficult to observe by light microscopy may be found with improved technique. A fuller report on these studies will be reported elsewhere.

REFERENCES

- Bredberg, G., Aden, H. W & Engström, H. 1972. Scanning electron microscopy of the normal and pathologically altered organ of Corti. *Acta Otolaryng* (Stockh.), Suppl. 501
 Engström, H., Aden, H W & Andersson, A. 1966. *Structural pattern of the organ of Corti* Almqvist & Wiksell, Stockholm.
 Engström, H & Engström, B. 1972. Structural and physiological features of the organ of Corti. *Audiology* 11 6.
 Hunter-Duvar L 1973 In preparation.

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Part 3 Effect on Cochlear Nuclei of Brain Stem (J G Hall)

The histological examination of the material was undertaken in order to find (1) whether the experimental lesions have produced degenerative changes in the neurons of the nuclei, and (2) if any loss of neurons demonstrable in these nuclei.

The total number of cells was computed in 10 of the cases according to the method de-

scribed elsewhere (Hall, 1964) For controls, 6 normal adult cats were also counted.

Two main types of neuronal changes were observed. One type was characterized by shrinkage of the perikaryon and pyknosis of the nucleus. The other type was distinguished by chromatolysis. Both types of degeneration occurred bilaterally on the labyrinthectomized

Table I Cell counts

R = Right side, noise-exposed L = Left side, labyrinthectomized

	Total cells		Ventral nucleus		Dorsal nucleus	
	R	L	R	L	R	L
\bar{X}	167 500	174 800	122 000	126 600	45 500	47 900
S	30 500	37 600	22 300	23 800	9 900	16 300
S _e	9 600	11 900	7 100	8 200	3 100	5 100
<i>Controls</i>						
\bar{X}	176 000		131 000		45 500	
S	15 300		9 400		7 200	
S _e	5 800		3 900		2 700	

\bar{X} the arithmetic mean; S, standard deviation S_e, standard error of the mean.

Table II. *Statistics*

Cat no.	Total cells		Ventral nucleus		Dorsal nucleus	
	Right	Left	Right	Left	Right	Left
23	152 800	142 600	112 500	105 200	35 400	37 400
24	155 800	205 500	114 900	162 000	40 800	43 600
25	171 800	200 800	131 900	152 900	41 200	52 800
26	129 900	92 400	92 400	72 100	32 400	18 600
27	182 300	208 300	125 700	132 000	61 600	76 300
28	156 800	145 400	122 900	113 000	42 200	38 400
29	125 300	169 700	87 200	118 300	40 700	36 800
30	216 500	201 900	156 500	141 200	53 100	62 200
31	213 000	188 900	152 800	134 300	60 200	54 600
32	170 500	192 400	122 800	134 500	47 800	57 900

as well as on the noise-exposed side of the animals. It was seen convincingly in the cochlear nuclei and also but to a much lesser degree, in other cell groups seen in the same preparations, for instance the superior olives or the cerebellar nuclei. The various groups of cells in the cochlear nuclei, e.g. the great and the small spherical cells, the pyramidal cells of the dorsal nucleus or the octopus cells, were affected to a varying degree but it was not possible from the microscopical examination to evaluate whether there was any difference in the reactions on the labyrinthectomized and the noise-exposed side. They both seemed equally affected. It was expected that a counting of the number of cells on each side would reveal some differences, either when compared with each other or with the normal material. The results of the countings showed that no significant difference was found between the number of cells in the operated cases and the controls (Tables I, II, III).

Furthermore, there was no significant differ-

ence between the two sides (labyrinthectomized and noise-exposed) neither regarding the total number of cells, nor the two main nuclei seen apart. Thus it may be stated that neither labyrinthectomy nor exposure to noise has induced any numerically demonstrable loss of neurons in the cochlear nuclei.

DISCUSSION

On the left side the degeneration of the nerve cells might possibly be due to the fact that their afferent fibres were cut and the hair cells destroyed. However both the efferent fibres following the bundle of Oort and the intrinsic fibres inside the nuclei may supply enough stimuli to prevent degeneration. This may account for the preservation of the cell population of the labyrinthectomized side. However we are also presented with degeneration of nerve cells induced after noise. This is more difficult to explain. Artifacts may give similar pictures, but in these cases all precautions were followed during perfusion, and normal cats undergoing the same infusion procedure did not show similar pictures. It may be that the findings are significant, and if so these cells would be functionally impaired, and induce a recruitment phenomenon through a break in the central pathways of the inhibition reflex. This pathway occupies three neurons, the cells of the spiral ganglion, the Octopus cells of Osen (1969) and the cells of the retro-olivary nucleus, possibly omitting the

Table III. *Controls*

	Total	Ventral nucleus	Dorsal nucleus
	195 600	141 700	54 000
	192 700	136 500	56 300
	164 900	121 300	43 700
R.	175 400	133 400	42 000
L.	151 200	115 200	36 100
R.	176 400	136 600	40 900
L.	177 500	132 000	45 500

efferent bundle of Rasmussen. If this constant inhibition should fail because of lesions of the pathway strong signals would be perceived unpimped, and a recruitment phenomenon might occur. Further investigations e.g. electronmicroscopy were suggested, whereby a more thorough examination of the nature of the cell changes might be possible.

REFERENCES

- Faro, M. D. & Windle, W. F. 1969 Transneuronal degeneration in brains of monkeys asphyxiated at birth. *Exp Neurol* 24 38.
- Hall, J. G. 1968 De centrale horelsbaners anatomi. *Nordisk Audiologi* 3-4 88.
- 1964 The cochlea and the cochlear nuclei in neonatal asphyxia. *Acta Otolaryng* (Stockh.), Suppl. 194 93.
- Hamberger C.-A. & Hyden, H. 1945 Cytochemical changes in the cochlear ganglion caused by acoustic stimulation and trauma. *Acta Otolaryng* (Stockh.), Suppl. 61 89.
- 1949 Production of nucleoproteins in the vestibular ganglion. *Acta Otolaryng* (Stockh.), Suppl. 75 53.
- Osen, K. K. 1969 Cytoarchitecture of the cochlear nuclei in the cat. *J Comp Neurol* 136 453.
- 1970 *The cochlear nuclei in the cat Felis Domesticus and the porpoise Phocoena Phocoena*. 51 pp. Universitetsforlaget, Oslo.
- Pfalz, R. & Pirsig, W. 1967 Neurale Regelung im Nucleus cochlearis durch negativen Feedback und deren Bedeutung für das zentrale Hören. *Hörheirliche-Akustik* 16 206.
- Powell, T. P. S. & Erulkar S. D. 1962 Transneuronal cell degeneration in the auditory relay nuclei of the cat. *J Anat* 96 249.
- Torvik, A. 1956 Transneuronal changes in the inferior olive and pontine nuclei in kittens. *J Neuro-path Exp Neurol* 15 119.

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EFFECTS OF NOISE UPON THE UPPER FREQUENCY LIMIT OF HEARING

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Abstract. The results of a thorough hearing examination in 228 young people indicate that the upper frequency limit is very sensitive to acoustic trauma, and that deviations from normal threshold are found more frequently above 5 000 Hz than at 3 000-4 000 Hz.

The Norwegian Broadcasting Corporation educates its own technicians and invites, each or every second year young people with examination perfect hearing and vision to apply for admission to the school. The selected students have a thorough audiological examination before they are finally accepted.

MATERIAL AND METHOD

Since 1954 I have performed this audiological examination, and since 1961 the testing and recording of data have been standardized. The material consists of 228 students from the classes 1962 to 1971 (201 males and 27 females) average age 20.7 years.

The testing was carried out in a sound-proof and anechoic chamber starting with an otoscopic examination. Detailed pure tone audiograms covering the frequency range 125 Hz-12 000 Hz were obtained by means of a Peters Audiometer (SPD 2 and AP 6) calibrated to ISO-threshold values.

Regular speech audiometry with monosyllabic numbers and PB-words was performed together with a randomized changing of inten-

sity level between 40 dB and 100 dB Loudness balance test (Fowler) examination of possible diplacusis binauralis, determination of difference limen for frequency and for intensity and measurement of the upper frequency limit were performed. A Radiometer HO 12 Oscillator and Tandberg HI FI Loudspeaker system 15 were used in the free-field listening tests.

The upper frequency limit was determined for one ear at a time, the listening ear turned towards the sound source. The tone, at a constant sound pressure level of 80 dB was continuously varied so as to cross the upper frequency limit in both an ascending and descending fashion. The SPL was determined using a Brüel & Kjær Precision Sound Level Meter at the place of the subject's ear after the subject had been removed.

RESULT

The observed sensorineural hearing losses were found in the frequency range above 2 000 Hz and were considered to be caused by exposure to high intensity sounds. According to the audiograms the material has been divided into 3 groups:

1 Deviations less than 10 dB between 125 Hz and 12 000 Hz: "Perfect Threshold" 113 subjects (50%).

2 Deviations less than 30 dB (and greater

Table I. Distribution of observed threshold deviations at various frequencies (in per cent)

Above 5 000 Hz only	80 (63)
At 3 000-4 000 Hz only	10 (5)
Both at 3 000-4 000 Hz and above 5 000 Hz	10 (32)

The numbers in parentheses are results from a similar investigation in 1962.

than 10 dB): "Small Deviations from Normal Threshold" 67 subjects (29%).

3 Deviations of 30 dB or more at one frequency or 25 dB at two adjacent frequencies: "Larger Deviations from Normal Threshold" 41 subjects (21%)

Of the 50 cases of noise-induced hearing loss (group 3) 34 were unilateral and 16 bilateral.

Examination of a possible difference between the male group having served in the military forces and the group without such service resulted in a higher percentage of noise induced hearing loss among male students being exposed to military noise—32% versus 18%.

The most frequently observed dips are localized in the frequency range above 5 000 Hz (Table I). The observed upper frequency limits are presented in Fig. 1. Almost half of group with "Perfect threshold" had an upper frequency limit above 17 000 Hz, whereas only 10% with Larger Deviations from Normal Threshold was able to hear frequencies above 17 000 Hz. Of this latter group 40% did not hear tones of frequency higher than 15 000 Hz, whereas only 1% of the group with Perfect Hearing had such a poor upper frequency limit.

The group with Small Deviations from Normal Threshold shows results lying between the other groups.

DISCUSSION

The findings in this selected material indicate that acoustic trauma causes threshold deviations

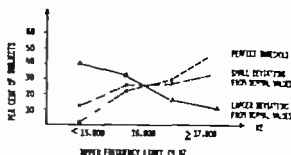


Fig. 1. Observed upper frequency limit as percentage of subjects in each group.

tions most frequently occurring above 5 000 Hz, and brings about a reduction of the upper frequency limit, even in cases with small deviations from normal threshold. Three different reasons are suggested as possible explanations:

1 Most ear defenders, most ear mufflers and some ear plugs have less attenuation above 5 000 Hz than between 2 000-4 000 Hz (own measurements).

2. Resonance effect of the ear canal (Wiener 1947, Djupesland & Zwislöck, 1972), effects of sound diffraction around the head (Wiener & Ross, 1946) and effects of the auricle under different sound incident angles (Berland & Nielsen 1968) result in increased pressure at the ear drum in the entire frequency range above 2 000 Hz, compared with the sound field with subject removed. Various resonance tops are seen in this higher frequency range.

3 The organ of Corti probably has poorer blood supply towards the base at least in some individuals (Siebenmann, 1894) and stria vascularis here appears poorly developed (Axelsson, 1968).

REFERENCES

- Axelsson, A. 1968. The vascular anatomy of the cochlea in man. *Acta Otolaryng* (Stockh.) Suppl. 243: 134.
- Berland, O. & Nielsen, T. E. 1968. Sound pressure generated in the human ear by a free sound field. Oticon Laboratories, Copenhagen, Denmark, 10 pp.
- Brødberg, G. 1967. The human cochlea during development and aging. *J Laryng* 81: 739.
- Djupesland, G. & Zwislöck, J. J. 1972. Sound pressure

- distribution in the outer ear *Acta Otolaryng* (Stockh.) 73 350.
- Steinmann, F. 1894. *Die Blutgefäße im Labyrinth des menschlichen Ohres*. Wiesbaden.
- Wiener F. M. 1947 On the diffraction of a progressive sound wave by the human head. *J Acoust Soc Amer* 19 143
- Wiener F. M. & Ross, D. A. 1946. Pressure distribution in the auditory canal in a progressive sound field. *J Acoust Soc Amer* 18 401
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BRIEF TONE AUDIOMETRY IN PATIENTS WITH ACOUSTIC TRAUMA

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Abstract Temporal integration of acoustic energy has been investigated in 25 persons with an acoustic trauma. The investigation showed a descending ability to integrate acoustic energy with increasing hearing loss for the frequencies 2 000, 4 000 and 8 000 Hz. These results were compared with results from a group of presbycusis patients, and it was shown that the relationship temporal integration/hearing loss is almost identical. A characteristic feature was normal temporal integration for some frequencies and abnormal temporal integration for others within the same ear. Taking the histopathology on noise-damaged cochleae into consideration, esp. localized hair cell degeneration, it seems a necessary condition that the outer hair cells are intact, in order to achieve a normal temporal integration.

The perception of brief tones is dependent on the stimulus duration in the following way: when halving the stimulus time, the sound energy must be doubled to maintain the threshold or to achieve equal loudness. This phenomenon is called temporal integration (TI) and is investigated by brief tone audiometry.

MATERIAL AND METHOD

Twenty-five patients are investigated. The diagnosis "acoustic trauma" is supported by case history and a typical audiogram.

The investigation is a monaural threshold determination performed in a sound-proof room. Each patient is tested at 3 or 4 of the frequencies 500, 1 000, 2 000, 4 000 and 8 000 Hz, and care has been taken to measure frequencies with normal as well as increased threshold in each ear. For each frequency in-

vestigated, ten threshold measurements are made using impulses of varying length from 1 000 msec to 1 msec. Procedure of investigation: rise-decay time, frequency characteristics of the impulses etc. are described earlier (Pedersen & Elberling, 1972).

The method of calculating the TI is illustrated in Fig. 1. The measured threshold relative to threshold for long tones are plotted in a double logarithmic coordinate system. A regression line for the plotted values is calculated and the area determined by X-axis, the Y-axis and the regression line is used as the relevant expression for the TI (Pedersen & Elberling, 1972).

RESULT

A typical record from one patient is illustrated in Fig. 1 and serves to demonstrate normal as well as abnormal TI within the same ear.

In Fig. 2 the TI at 4 000 Hz is correlated to the actual hearing loss. The figure shows that the TI decreases when the hearing loss increases. Same results were found at 2 000 and 8 000 Hz. In Fig. 2 the results at 4 000 Hz from patients with acoustic trauma are compared with the results from 46 presbycusis patients.

DISCUSSION

It has been demonstrated that the TI is reduced in patients with acoustic trauma, and

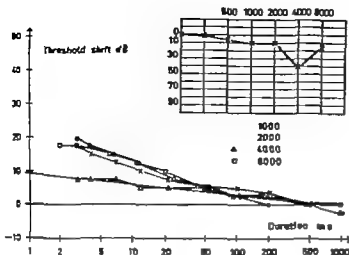


Fig 1 Brief tone audiometry at the frequencies 1 000 2 000 4 000 and 8 000 Hz in a case of acoustic trauma. Audiogram upper right. The fig. shows normal temporal integration at the frequencies 1 000, 2 000 and 8 000 Hz, while the temporal integration is small at 4 000 Hz (14 normal value 28).

that the threshold shift obtained when using a long and a short tone impulse decreases, when hearing loss increases. The change in T.I. for a given hearing loss was almost the same as found in a group of presbycusis patients (Pedersen & Elberling, 1973).

Degeneration of outer hair cells and nerve fibres in the cochlea is the characteristic finding in animal experiments on acoustic trauma. The localization and degree of degeneration

being dependent on stimulation time, frequency and intensity (Engström et al., 1970).

In human cochlea too it has been possible to demonstrate a relation between acoustic trauma and localized hair cell degeneration (Bredberg, 1968).

The results of brief tone audiometry on persons with acoustic trauma (Fig. 1), showed normal and abnormal T.I. for different frequencies, within the same ear. Comparing these results with the localized hair cell degeneration in acoustic trauma patients, it seems reasonable to conclude that intact outer hair cells are necessary for normal T.I. of acoustic energy.

REFERENCES

- Bredberg, G. 1968. *Hearing mechanisms in vertebrates*. Churchill, London (Ciba Symposium).
 Engström, H., Aden, H. W. & Bredberg, G. 1970. *Sensorineural hearing loss*. Churchill, London (Ciba Symposium).
 Pedersen, C. B. & Elberling, C. 1972. Temporal integration of acoustic energy in normal hearing persons. *Acta Otolaryng (Stockh.)* 74: 398.
 — 1973. Temporal integration of acoustic energy in patients with presbycusis. *Acta Otolaryng (Stockh.)* 75: 32.

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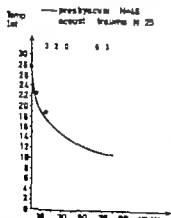


Fig 2. Temporal integration correlated to the hearing loss at 4 000 Hz for patients with acoustic trauma. The values are pooled into 10 dB groups. Number of cases in each group are indicated. — mean values of temporal integration in 46 patients with presbycusis.

BINAURAL DISCRIMINATION OF "EVERYDAY" SPEECH

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Abstract The binaural intelligibility of Pb-words in 9 indoor everyday listening situations is tested on 63 persons with bilateral sensori-neural losses and 22 normals. A group with bilateral sensori-neural loss of 50 dB or more on 3 000 Hz and above and normal hearing up to and including 2 000 Hz have 25-45% lesser scores than normals. The corresponding figures for bilateral normal hearing up to 3 000 Hz and bilateral sensori-neural loss of 50 dB or more on 4 000 Hz and above is 5-15%. In quiet there is about 5% difference between normals and these groups.

Kryter et al. (1962) and Harris (1965) have shown that the three most important frequencies in the tone-audiogram for estimating speech intelligibility of English Pb-words and sentences in "everyday situations" are 1 000, 2 000 and 3 000 Hz. In order to investigate the

importance of a hearing loss of frequencies over 2 000 Hz in such situations, nine different simulated listening situations in "everyday milieu" have been recorded. The recording room was the sitting-room of a three-room flat. This had an area of 20 m², a volume of 50 m³ and a reverberation time of 0.5 seconds. The recordings have taken place over an artificial head without auditory canals. The speech signal, noise and competing speech have been presented via separate loudspeakers. In situations 7-9 (see below), the competing speakers have been authentic, and only the speech signal has been presented via loudspeaker. The following listening situations have been recorded.

- 1 Signal/noise = 0 dB
- 2 Signal/noise = -5 dB
- 3 Signal/noise = -10 dB
- 4 Signal/noise = 0 dB + competing radio voice
- 5 Signal/noise = -5 dB + competing radio voice
- 6 Signal/noise = -10 dB + competing radio voice
- 7 Signal + 1 competing speaker
- 8 Signal + 2 competing speakers
- 9 Signal + 3 competing speakers

Distance, speaker to listener = 3 m.

Distance between all speakers
(incl. signal) and listener = 1 m.

Recordings 1-6 simulate a listening situation with and without competing radio voice and a traffic noise level of 63-73 dB (A) outside the window. If the latter is of normal construction. In newly built houses there are windows where the outdoor level would correspond to 68-78 dB (A). The playbacks have taken place at the same level as the recordings and have been presented to 22 normal-hearing persons and

63 with sensori-neural loss, via binaural headphones. The hard-of-hearing persons have been divided into groups. The two most interesting of these consists of a group L 3 000 of 22 persons with normal hearing up to and including 2 000 Hz and thereafter binaural hearing loss of 50 dB or more at 3 000, 4 000, 6 000 and 8 000 Hz and a group L 4 000 of 6 persons with normal hearing up to and including

3 000 Hz and thereafter 50 dB hearing loss or more at frequencies 4 000, 6 000 and 8 000 Hz. In a comparison between these groups and the normal hearing persons, one finds that the binaural speech intelligibility in a quiet milieu, for normal hearing as well as for the two groups with hearing loss, is 90-95%.

The listening situation signal/noise = 0 dB + competing radio voice simulates a fairly normal background noise + 1 competing voice in a room with closed windows in an urban residential area. One finds in this situation that group L 3 000 with hearing loss of frequency 3 000 and above has 30-40% speech intelligibility while the normal hearing persons have 70-75%.

Group L 4 000 with normal hearing up to and including 3 000 Hz, has in this situation hardly 60%. If one converts the speech comprehension percentage for Pb-words into a corresponding one for sentences, one finds that the group L 3 000 misunderstands every third sentence in this listening situation while the normal hearing group only misunderstands every twentieth. In the situation with 1 speaker and 3 competing speakers, which simulates an everyday situation which is particularly common, one finds that group L 3 000 has 25-30% speech intelligibility of Pb-words while normal hearing have 65%. Group L 4 000 has 50%. Converted into sentences, this means that group L 3 000 misunderstands every second sentence, while a normal-hearing only misunderstands one of 20. A comparison between the performance of normal hearing and that of so-called "slightly hard of hearing" persons in more or less difficult listening situations, which are daily typical for the greater part of the population, shows that one attaches far too little importance to hearing losses of frequencies over 2 000. This is due to the fact that speech intelligibility has hitherto chiefly been measured in quiet surroundings, which when it concerns diagnostic tests is naturally necessary. When it is for the purpose of getting a grasp of the handicap of a hard-of-hearing person in "everyday milieu" however judging

by the result of this present work, it is hardly relevant. The author therefore suggests that the battery of tests which the otologist of today has at his disposal, should be further increased by one, namely speech intelligibility in "every day milieu".

A correlation analysis has been carried out, whereby those recordings which have given the greatest correlation to hearing loss, according to the tone-audiogram have been chosen. Comparison has been made between listening with headphones and in sound field which has shown that headphone listening gives as good or somewhat better results than sound field listening. The hard-of-hearing groups and the normal group with which they were compared, have gone through a lesser verbal test which shows that the vocabulary of the test persons, has not affected the test result.

Comparison between a group who have retained information on frequency 3 000 in only one ear while overlying frequencies have a hearing loss of 50 dB or more (L 3 000) and the groups L 3 000 and L 4 000 (50 dB or more bilaterally on frequencies 3 000 resp. 4 000 and above) has been carried out. This shows that speech intelligibility is fundamentally dependent on the better ear in a listening situation where there is a diffuse noise while an asymmetric hard-of-hearing person needs two good ears to a higher degree in a listening situation like that of signal + 3 competing speakers.

Eight groups of 7-10 normal-hearing medical students (a total of 64) have been tested in groups, each on a filtered version of three of the above mentioned recordings, i.e. signal/noise = 0 dB, signal/noise = 0 dB + radio and signal + 3 competing speakers. The result of this investigation shows that a falling off of frequencies over 2 500 Hz gives as great a loss of speech intelligibility as a falling off of frequencies below 1 000 Hz and that frequencies over 3 200 Hz are just as important as those below 700 Hz.

A comparison has been made between normal hearing who were tested on low-pass fil-

tered speech (artificial hearing loss) and persons with corresponding sensori-neural loss. The result shows that the persons with hearing loss did not get better results than the corresponding artificially injured groups. To sum up the investigations show that one should allot great hearing losses on frequencies 3 000 and 4 000 Hz, just as much importance as great losses on frequencies 500 and 1 000 Hz, when estimating a person's speech intelligibility in "everyday milieu".

French & Steinberg (1949) developed a method whereby one could calculate from purely physical measurements, an index to the intelligibility of speech. They called this the Articulation Index or AI. AI calculations have been carried out for listening situation $S/N = 0$ dB, in unfiltered version, and in 7 filtered versions, and for listening situation $S/N = -5$ dB and $S/N = -10$ dB. Comparison has been made, between the expected result applicable to a vocabulary of 1 000 English Pb-words and the results received in the investigation. It shows that our results agree well with those expected, in spite of the material being Swedish Pb-words. This supports

the view that a Swedish Articulation Index would not differ materially from an English one. Corresponding AI calculations have also been made for the hard-of-hearing groups and the results show that the expected speech intelligibility according to a vocabulary of 1 000 English Pb-words agrees closely with the actual result of our investigations. This means that AI calculations for persons with sensori-neural loss on frequencies over 2 000 Hz listening on speech in diffuse noise is a good method for estimating speech intelligibility.

REFERENCES

- French, N. R. & Steinberg, J. C. 1949. Factors governing the intelligibility of speech sounds. *JASA* 19, 90.
 Harris, J. D. 1963. Pure-tone acuity and the intelligibility of everyday speech. *JASA* 37, 824.
 Kryter, K. D., Williams, C. & Green, D. M. 1961. Auditory acuity and the perception of speech. *JASA* 34, 1217.

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COMMUNITY DIAGNOSIS OF PROFESSIONAL NOISE TRAUMA

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Abstract. 92 367 persons in all of Denmark applied for hearing rehabilitation in a 5-year-period, and of those 7 108 (7.7%) were registered under the diagnosis professional noise-induced hearing loss. I.e. an incidence of about 1 400 per year or 28 per 100 000. 40% were below 65 years of age, and only 6% were women. There are estimated to be 6 000 industrial works employing about 50 000 persons at present with a continual noise level of more than 90 dB (A). A hearing conservation program has been proposed to The Ministry of Work.

Denmark has a population of 4.9 mill. inhabitants, who are carefully registered from birth until death. Since 1951 every Danish citizen with permanent hearing impairment may have complete rehabilitation free of charge including 1 or 2 modern hearing aids from the current market. Therefore very few people buy a hearing aid of their own, and thus the group of hearing handicapped persons passing through the hearing clinics indicates the morbidity nearly 100%. The hearing aids are supposed to last for at least 5 years, concerning adults, and only in exceptional cases are the patients examined more than once in a period of 5 years.

METHOD

Information was collected from the three state hearing rehabilitation centres and their nine daughter clinics concerning the morbidity of clinical hearing impairment. To even out yearly variations, data were collected over a 5 year period during the years 1964-1970. The criteria for the diagnosis degenerative nervi

acustici professionals are: normal otoscopy nearly symmetrical hearing loss on both ears with a maximum around 4 000 or 6 000 Hz and a case history that the subjects have been exposed to relatively long periods of loud industrial noise, in which speech at a normal level could not be perceived at a short distance.

RESULT

In the 5 year period 92 367 patients who felt permanently hard of hearing applied for an audiological examination and rehabilitation. Among those 7 108 = 7.7% are registered under the diagnosis: degenerative nervi acustici professionals. The important fact of this is that about 1 400 new patients per year are applying for audiological treatment, because they feel hard of hearing in their daily life due to noise exposure, i.e. 28 per 100 000 inhabitants. More detailed information has been gained from three hearing clinics, to which 16 671 patients applied because of a permanent hearing disorder. 1 108 patients are registered as chronic noise trauma (6.6%). Of those, only 6% were women, and 60% of the patients with noise induced hearing losses were over 65 years of age. In Table I the 1 108 patients are grouped according to their speech reception threshold. 200 had a normal speech reception threshold, although they had a noise dip. Half of the patients are registered with a slight impairment, 300 as medium, 80 as severe and finally 23 as deaf. From the case records of the

Table I *Severity of noise trauma indicated by Speech Reception Threshold*

<i>Normal hearing</i>	
SRT 0-20 dB ~	200 persons (18.0 %)
<i>Slight impairment</i>	
SRT 25-40 dB ~	505 persons (45.6 %)
<i>Medium impairment</i>	
SRT 45-60 dB ~	300 persons (27.1 %)
<i>Severe impairment</i>	
SRT 65-80 dB ~	80 persons (7.2 %)
<i>Deaf</i>	
SRT >85 dB ~	23 persons (2.1 %)
Total	= 1108 persons

tients it is quite obvious that some patients get a severe noise trauma after a few years exposure, while other patients maintain nearly normal hearing regardless of 20-30 years exposure to heavy noise.

Noisy Industries in Denmark

Only about 25 factories have more than 1 000 workmen. The great majority of Danish industries have less than 25 employees. On one large island (Fyn) with around 700 000 inhabitants the representation of various kinds of industries is typical for Denmark. This area has been carefully explored, and from the results we estimate that we have, in Denmark as a whole 6 000 enterprises with a continuous noise level of 90 dB (A) or more. In these enterprises about 50 000 workmen are employed. It is calculated that the noise level could be reduced to less than 90 dB (A) by means of moderate expenditure in approximately 80 of these factories. In the remaining works in which noise cannot be reduced below the risk level the employees must make use of ear protectors, which are regarded as a last resource. Engineers and medical audiologists have proposed to The Ministry of Work a detailed hearing conservation program with

education of audio-technicians who should regularly perform audiometric examinations in industries. The effort should be concentrated around every new person who is going to be employed in industry with heavy noise. These new persons should have taken their pure tone audiogram in course of the first weeks of the employment. Then they should be followed with an interval of half a year with a control of the air conduction curve only in respect of the frequencies 2 000, 4 000, 6 000 and 8 000 Hz. If the hearing deteriorates more than 10 dB the employee should consider being placed in a more quiet part of the enterprise.

DISCUSSION

The influence of noise upon human beings is dependent on the noise level, the noise spectrum, the duration and the individual's resistance to noise. Like other sense organs, hearing is subject to a certain degeneration dependent of age. In a person with a noise-induced hearing loss this loss will be added to the hearing loss of aging. It is therefore usual that people with occupational hearing losses get along quite well through their thirties and forties, until they come to the age of 50. Then they begin to feel their hearing handicap more and more. This means that the noise wears out the reserves 10-20 years earlier than we would have expected due to the aging process. Most workmen ignore this happening. Therefore we have the task which is necessary to obtain the noise level which we want, and it is important that not only the physicians but all audiologists and the employed persons themselves are active in the abatement of harmful noise.

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A METHOD FOR COMPUTERIZED CLASSIFICATION OF PURE TONE SCREENING AUDIOMETRY RESULTS IN NOISE-EXPOSED GROUPS

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Abstract Screening audiometry data from noise-exposed groups are automatically processed by a computer. The process results in a two digit number for each individual showing the hearing situation for each ear. The method has several advantages in that it is simple, rapid and inexpensive.

Pure tone screening audiometry has become widely used as an important element of the hearing conservation programs for noise-exposed personnel. Usually only hearing losses exceeding a certain level (e.g. 15 dB) are recorded at selected frequencies (e.g. 500 1000 2000 3000 4000, and 6000 Hz).

This method is rapid and simple enough for non-specialized investigators to perform. Since there is a frequent occurrence of hearing defects in noise-exposed groups, civil and military health service programs reveal a variety of pathological conditions. Consequently there is an increasing demand for a rapid and uniform evaluation method to provide the basis for preventative measures.

The new method entails computerized classification based on reading of pencil marks from a new "Optical Mark Scoring Sheet" used during the screening process. This sheet has sections for marks of personal identification, hearing loss for each ear at the frequencies mentioned above, and for the oto-

logical case history with particular reference to noise exposure. The sheets are fed into an "Optical Mark Page Reader" connected to a computer (IBM 370/155 at the Uppsala University Data Centre). The marks denoting the hearing are converted by the computer into an individual two-digit number the first digit of which refers to the right ear and the second to the left.

The digits are from 1-5 depending on the extent to which the hearing losses involve certain areas of a pure tone audiogram sheet shown in Fig. 1. As an example, a two-digit number of 24 indicates "slight high tone loss" in the right ear and "severe high tone loss" in the left. A number of 35 indicates "moderate high tone loss" in the right ear and "other hearing loss" in the left, the latter being of unknown type until further investigation has revealed its character.

The two-digit number provides a fairly good view of the individual hearing situation especially in asymmetrical findings. First, a rough idea of the social hearing ability may be concluded from the lesser digit which refers to the better ear which is of dominant importance for the social hearing. Secondly the greater of the two digits, in case of asymmetrical hearing loss, indicates a hearing defect which is probably not caused by continuous acoustic trauma alone.

Previous methods have involved manual procedures and evaluations in which the da

This project is sponsored by the National Swedish Council of Building Research and the Swedish Building Industry Work Research Foundation (Project 1310 BAS).

HEARING CLASSIFICATION OF NOISE EXPOSED PERSONNEL

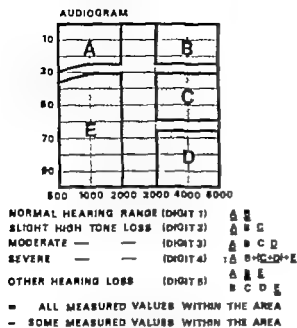


Fig 1 The audiometric criteria according to which the marks denoting hearing loss are converted by the computer into a digit 1-5 for each ear

of both ears have been weighed together into one single expression (Heijbel & Lidén, 1957

Lindqvist, 1970) This may be insufficient in cases of asymmetrical defects of differing character. The new method of classification has the additional advantage of being fully automated. The optical mark scoring sheets used at screening can be mailed directly to the data centre and the classification results are rapidly obtained. The results may be stored for follow-up and for additional statistical processing of any desirable aspect. With the aid of a short instruction manual the need for otologic specialist consultation may generally be limited to some of the cases in which digit 4 or 5 occurs.

Finally the method is very inexpensive as compared to other approaches to the problem.

REFERENCES

- Heijbel, C. A. & Lidén, B. 1957 Klassificeringssystem för hörselskador Nord Med 58 959
Lindqvist, S. 1970 Hörselskador hos byggschaktare, Svensk Läkartidn 67 4283

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A HEARING CONSERVATION PROGRAMME FOR SMALL INDUSTRIES IN A SWEDISH COUNTY

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Abstract Östergötland's County Council has organized an industrial hearing conservation programme mainly aiming at small industries where knowledge and own resources presumably are small. At a subsidized cost the programme offers noise measurements, screening audiometry information on noise control and ear protectors and a full audiological examination of certain cases. During 1971 we visited 65 companies. The results of screening audiometry (3 500 cases) and audiological examination of 212 cases are presented and discussed.

During recent years a growing need has been felt for studying industrial noise and preventing its damaging effects on exposed people. Although not sufficient, considerable activity has been going on for several years in Sweden, e.g. through health services organized by some, mainly larger companies, through the Labour Inspectorate, through some industry oriented projects, etc. In our early sporadic attempts in this area we found a great need for a permanent organization, preferably with a close medical connection. The problem is essentially one of information—informing the employer that improvement in noise levels in the long run is beneficial for production and means less risk of hearing loss and less discomfort for his employees, informing the employee that in too high noise levels not only his neighbour's but also his own hearing is at risk, that signal-to-noise ratio is sometimes better with ear protectors than without.

This permanent organization was established in 1971 as a part of the Department of Audiology at the Regional Hospital, Linköping. A special committee was organized, representing

all the groups that have any interest in the programme (County Council, hospital, Labour Protectorate, labour unions, employers) to determine the general plans and rules of the programme. The staff consists of one electrical engineer one audiometrist and a secretary plus a consulting engineer equipped with a bus with a small sound-insulating box for audiometry audiometer sound level meter tape-recorder for recording transient sounds and an assortment of different types of ear protectors.

We have access to the company-register of the district office of the Labour Protectorate and contact mainly small firms (up to 100 employees) in risk-industries (metal, wood, printing, chemical, textile, food, stone). We offer our noise-programme at a subsidized cost (approx. one-quarter of the real cost). During 1971 we contacted ca. 200 small industries by mail, of which 23% wanted to be included in the programme. Another 12% could be persuaded adding up to 35% positive, 36% negative and 29% offering no response at all to our letter. This must be interpreted as a rather moderate interest from the employers' side in our programme, probably partly due to ignorance of noise as a problem, partly due to economic considerations—not so much out of a fear of the cost of this programme, as of the demand for an improved working environment that might follow.

Sixty-five companies were visited noise and hearing measurements performed and informa-

tion given on noise reduction and ear protectors.

After analysis of the noise measurements and application of risk criteria a written report is sent to the company. Audiograms are classified in five categories according to a rather simple system suggested by Drettner et al. (1970). Before audiometry everybody is instructed to fill in a questionnaire, giving some personal and anamnestic data. The results of the audiometric classification together with these questionnaires are then studied in order to select those who ought to be subjected to a full audiological examination. A report on this classification and selection is sent to the company including a special letter of information to those who were found to have a slight, moderate or moderately severe hearing loss, presumably due to noise exposure. Based on the screening audiometry of the total material (3 543 cases) 32.5% were found to have bilateral hearing thresholds which were classified as normal (maximum 35 dB at 250 Hz, 30 dB at 500 Hz and 25 dB at 1 000–6 000 Hz) while ca. 8% were classified as bilateral severe hearing loss, presumably noise induced (severe loss defined by threshold exceeding above mentioned limit at some but not all frequencies in the range 250–2 000 Hz in on to the high frequency loss).

The group selected for further examination consists of three subgroups: (A) Cases with considerable asymmetry in the audiograms. (B) Cases with severe loss presumably due to noise, particularly those of young age. (C) Cases whose audiograms indicate other types of hearing loss than noise-induced, previously undiagnosed.

Out of the 3 543 audiograms obtained in 1971 212 cases (6%) were selected for further examination. Category A made up 27 cases, B 60 and C 125 cases. In 8 of the cases in category A the most likely diagnosis remained noise-induced hearing loss after audiological examination while in category B this was true for 48 cases. The original preliminary diagnosis of noise-induced loss for the re-

maining 19+12 cases (35%) proved to be incorrect. In category C, 39% had a conductive hearing loss and 40% a sensori-neural loss most likely not caused by noise exposure. Six of the conductive cases have accepted offers of middle-ear-surgery. Eight cases have obtained hearing aids and 14 cases other technical aids for the hard-of-hearing.

In this selected group a comparison has been made between hearing thresholds obtained in the clinic and in our field-bus. The mean differences and standard deviations obtained were -2.1 ± 9.8 dB at 250 Hz, -0.9 ± 8.1 dB at 1 000 Hz and -0.9 ± 7.2 dB at 4 000 Hz with clinical thresholds in average better. The standard deviations are somewhat larger than might be expected (Delany 1971) which may be explained by TTS at the field-audiometry by a disproportionately large number (10%) with temporary loss due to upper respiratory infections in the selected group, untrained and unsophisticated listeners, etc.

A check-up has been done on the accuracy of the manual use of the classification system for audiograms. Out of 500 randomly selected audiograms, 3% had been wrongly classified.

A system for the classification of audiograms is valuable since it offers a relatively clear view of the status of hearing in a population. If the number of classes is small and clearly defined audiometrically it will also be quite simple to use manually. However if the classification is done automatically e.g. by a computer simplicity is no longer an important advantage, and the diagnostic accuracy should be improved by using a more sophisticated system.

REFERENCES

- Delany M. E. 1971. In *Occupational hearing loss* (ed. Roblason). Academic Press, London and New York.
Drettner B., Klockhoff I. & Lindholm, L. 1970. Personal communication.

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PROPHYLACTIC MEASURES AGAINST INDUSTRIAL NOISE

A Fifteen year Survey of the Work done in a Swedish County

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Abstract. During a 15-year period there has been a successive and successful prophylactic work against industrial noise in the county of Örebro, Sweden. Medical as well as technical advice is given to the industries; informative courses are given, too.

The building up of small hearing centres in the various counties of Sweden started in the 1950s. One of the first of these centres was in the county of Örebro, the population of which is about 280 000.

To start with basic audiometry was performed, but the major aim of the hearing centre was to channel the prescription and the selling of hearing aids in a firm and ethically correct way. The senior physician of the ENT-department, two so-called "hearing assistants" and one audiometrist were responsible during the first years for the hearing rehabilitation program. Later a social worker was attached to the hearing centre.

As there was a great interest in the county council of Örebro in hearing rehabilitation and in hearing prophylaxis some screening investigations were also undertaken. The most natural one was—as in other hearing centres of Sweden—the screening made in ordinary schools, but in Örebro there also started a screening of the hearing of the workers at the Central Workshops of Swedish Railways. For various reasons, however the investigations were not carried out on a continual basis, though in 1957 a technician was employed at the hearing centre. He was expected to do

some repair work on hearing aids and diagnostic equipment, and to start a hearing investigation program for the noisy industries.

Only one factor in the county had at that time a good prophylactic hearing program—Bofors AB. The other factories, small as well as larger ones, had only a summary survey of their noisy conditions, made by the Yrkesinspektion (the Swedish Factory Inspectorate).

The hearing centre offered its equipment and technical assistance to the firms, and medical advice was also included in this. After the investigations of the noisy parts of the factory were completed, attention was turned to the individual worker and his hearing. All those working in more than 85 dB (A) were to be examined at least once a year. Information and advice was to be given. All the costs for the hearing centre were met by the firm itself.

A very slow and modest start was a good help to consolidate the work, obtain experience and make improvements. A good deal of the early work was propagandistic, i.e., to inform the firms and the workers about the possibilities in this part of the health program.

After some years there was a stand-still until in 1966 an engineer with the sole task of dealing with the acoustic problems, especially industrial ones, joined the staff of the hearing centre. In fact, he turned out to be the first one in Sweden employed by a county council at a hearing centre to combat noise. Since

tion given on noise reduction and ear protectors.

After analysis of the noise measurements and application of risk criteria a written report is sent to the company. Audiograms are classified in five categories according to a rather simple system suggested by Drettner et al. (1970). Before audiometry everybody is instructed to fill in a questionnaire, giving some personal and anamnestic data. The results of the audiometric classification together with these questionnaires are then studied in order to select those who ought to be subjected to a full audiological examination. A report on this classification and selection is sent to the company including a special letter of information to those who were found to have a slight, moderate or moderately severe hearing loss, presumably due to noise exposure. Based on the screening audiometry of the total material (3 543 cases) 32.5% were found to have bilateral hearing thresholds which were classified as normal (maximum 35 dB at 250 Hz, 30 dB at 500 Hz and 25 dB at 1 000-6 000 Hz), while ca. 8% were classified as bilateral severe hearing loss, presumably noise induced (severe loss defined by threshold exceeding above mentioned limit at some but not all frequencies in the range 250-2 000 Hz in addition to the high frequency loss).

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REFERENCES

- Delany M. E. 1971 In *Occupational hearing loss* (ed. Robinson). Academic Press, London and New York.
Drettner B, Klockhoff I & Lindholm L. 1970. Personal communication.

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MUSIC—A NOISE HAZARD?

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Abstract. Measurements of music noise spectra in various youth centres show great variations in levels in rooms with different acoustic properties. The noise spectra from different orchestras are, however remarkably equal. The high level and the narrow dynamic range, together with the likelihood of being very close to the loudspeakers give reason to believe that the criterion of damage risk is surpassed by listeners and musicians rather frequently. Actual hearing measurements seem to confirm this suspicion.

During the last 10 years not only has unwanted environmental sound increased in intensity but also recreational sound, such as the music played by "pop groups" and performed in discotheques.

Although most measurements of such musical sounds seem to indicate intensity levels above or on the borderline of damage risk criteria, the observations of "music-traumatic" hearing losses vary from place to place. Ewertsen (1971) concludes that in Copenhagen no cases of deafness caused by "pop music" have been seen among 80 000 individuals with hearing loss examined.

Lipscomb (1970) found a significant increase in hearing loss in the frequency range above 3 000 Hz among college students and concluded the findings to be tangible evidence of the toll being exacted by "pop music" sounds.

Speaks et al. (1970) having measured music noise spectra and temporary threshold shifts summarize their results: the exposures are "borderline"

My investigation of the hearing status of

broadcasting technicians starting their training (Flottorp 1972) indicates that incidence of acoustic-traumatic hearing loss among young people has increased during the last 10 years. In order to examine whether a possible causality exists between this observation and the exposure young people get to music noise in youth centres, I have measured the sound level in several such centres in the Oslo area.

APPARATUS AND METHOD

The measurements were carried out with a Precision Sound Level Meter (Brüel & Kjær type 2201) calibrated with a Pistonphone (Brüel & Kjær type 1613). Sound levels were measured at several locations on the general dancing area and at restaurant tables, as close to the loudspeakers as listeners used to come and at more distant places.

Usually a frequency analysis in octave bands was done, though a number of measurements were made using the A-weighting network.

The "slow" response was mainly used, however some measurements of drum-noises were taken using the "fast" response. An estimated average of frequent peaks observed on the meter was recorded.

RESULT

The results for some orchestras are presented in Figs. 1-2. There is remarkably good agree-

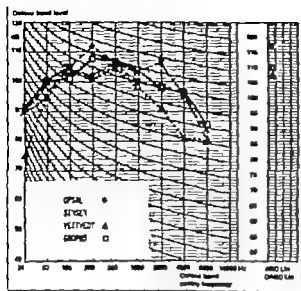


Fig 1 "Music noise" in various youth centres in Oslo.

ment between noise spectra observed for different orchestras. Thus, the spectra reported here are in good agreement with those reported by Lebo et al. (1967) Rintelmann & Borus (1968) and by Speaks et al. (1970).

My results show however even more energy in the highest frequency range in some instances.

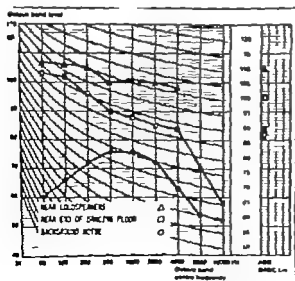


Fig 2 "Music noise" at various distances from the orchestra "Beatniks"

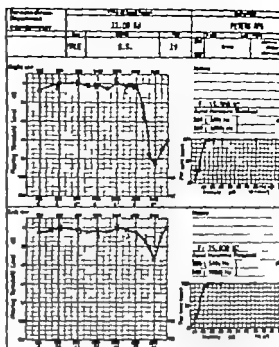


Fig 3 Hearing loss in a rock-and-roll musician.

The sound pressure level in octaves above 4 000 Hz depend very much upon the distance from the loudspeakers, because the crowd of young people absorbs much of the energy in the highest frequency range.

In the rear part of the dancing floor the sound spectrum is completely different from that in the front part, as shown in Fig. 2.

The reverberation time of the actual hall also is an important factor in the resulting spectrum and sound pressure level at the listener's place. Very often, basements are used for youth clubs, and here the reverberation time may amount to 3-4 seconds. It is not unusual that such basements are used by schools for training and rehearsals by the school's brass band. The sound level in such rooms is definitely higher than when the same band performs in an acoustically treated room or a larger room.

Quite a few youth clubs and discotheque have rather short reverberation times, and the sound level of the music performed there has been measured to be definitely lower than the results presented in this paper.

DISCUSSION

A typical feature of modern "pop music" is its very narrow dynamic range. When the group starts, it immediately exceeds the normal maximum level of a symphony orchestra, and keeps this level within ± 10 dB. This makes "pop music" much more hazardous to the hearing than music performed by great symphony orchestras.

Hi-fi amplification systems, with amplifiers yielding several hundred Watts, are used by the groups. Very often the loudspeakers are placed only 1 metre from the listeners, resulting in a sound pressure level at the listener's ear of 120–130 dB.

The loudspeakers are placed in front of the performing musicians. Very often however the hall and especially the stage accommodated by the musicians is rather reverberant, causing very high levels at the ears of the musicians also. Many of the listeners are exposed to this type of music noise 2–3 hours almost each day and the musicians more than 6–8 hours a day.

Taking into account the possibility of getting very close to the loudspeakers and also the fact that rooms used for this type of music very often are too reverberant, I believe the borderline of damage risk criterion is probably

surpassed by listeners and musicians rather frequently. The reported increase in noise induced threshold deviations (Flottorp, 1972) seems to confirm such a suspicion. An audiogram of a member in a "pop group"—not otherwise noise exposed—is shown in Fig. 3 as an indication of the hearing hazard of "pop music".

REFERENCES

- Evertsen, H. W. 1971. Beat music and damage to hearing. *Nord Audiol* 20: 154.
 Flottorp, G. 1972. Effects of noise upon the upper frequency limit of hearing. *Acta Otolaryng* (Stockh.) 75: 329.
 Lebo, C. P., Oliphant, J. S. & Garrett, J. 1967. Acoustic trauma from rock-and-roll music. *Can J Med* 107: 378.
 Lipscomb, D. M. 1970. The increase in prevalence of high frequency hearing impairment among college students. *Abstr X Int Congr Audiol* 99.
 Rintelmann, W. F. & Borus, J. P. 1968. Noise-induced hearing loss and rock-and-roll music. *Arch Otolaryng* (Chic.) 88: 377.
 Speake, C., Nelson, B. & Ward, W. D. 1970. Hearing loss in rock-and-roll musicians. *J Occup Med* 12: 216.

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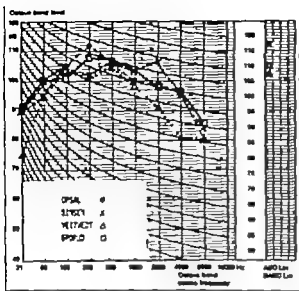


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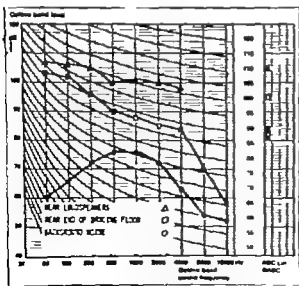


Fig 2 "Music noise" at various distances from the orchestra "Beatniks".

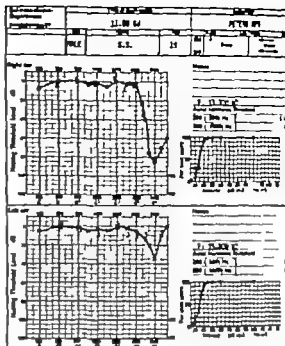


Fig 3 Hearing loss in a rock-and-roll musician.

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Quite a few youth clubs and discotheques have rather short reverberation times, and the sound level of the music performed there has been measured to be definitely lower than the results presented in this paper.

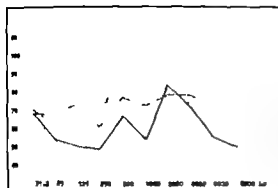


Fig 1 Signal of four different emergency vehicles. Measured with octave filter set.

cars, hearing at 60 dB level, or when using cotton prosthesis, at 30 dB was used as a handicapped subject. Generally the hearing-handicapped subject heard the warning signals of emergency vehicle 10–30 m before or as quickly as the normal persons. The worse the attenuative properties of car bodies were, the better the hearing-handicapped subject was able to observe the warning signals.

Is there discrimination against hearing-handicapped persons in traffic? Would it not be better to take as criterion ear diseases rather

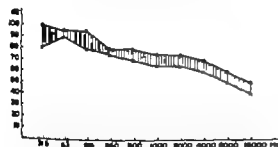


Fig 2. Traffic noise in street corner. Total noise 76–92 dB (A). Measured with octave filter set. Average ○ maximal level.

Table I. Attenuative properties of bodies of four cars in dB (A)

	Sound source		
	Front	45° angle	90° angle
Bedford (truck)	21 dB (A)	25 dB (A)	19 dB (A)
Renault R4	28	29	25
Saab 99	33	38	34
Toyota Crown	52	55	47

Table II. Subjective loudness of alarm signal in traffic conditions

Warning signal heard	Distance of emergency vehicle from street corner			
	25 m	50 m	75 m	Total
At a distance before street corner	3	6	2	11
Immediately before street corner	14	7	10	31
In junction box	18	12	15	45
After street corner	1	3	9	13
Not heard	1	3	3	7
Total	37	31	39	107

41 drivers could not localize the emergency vehicle.

than hearing ability in the issuing of driving licences? One can ask too, is there discrimination against all drivers other than those of emergency vehicles? According to traffic law an emergency vehicle possesses the right of way. Is a driver unprotected in court in a case involving collision with an emergency vehicle?

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SOUND PRESSURE DISTRIBUTION IN THE OUTER EAR

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Abstract Sound pressure transformation between several points in the outer ear and the eardrum was measured by means of a probe-tube microphone. The tip of the probe tube was positioned with the help of a fine screw arrangement and a Zeiss surgical microscope. The results slightly amend and extend the classical data of Wiener and Ross.

It is known that the sound pressure at the entrance of the ear canal is not the same as the sound pressure at the eardrum except at very low audible frequencies. The variation of sound pressure along the human auditory canal was measured in a classical study by Wiener & Ross (1946) with the help of a small probe-tube microphone. The sound pressure distribution in the ear canal has been computed theoretically by means of an electrical network model (Zwislocki 1965). Small discrepancies have been found between the analog results and the experimental results of Wiener and Ross. In order to find the reason for the differences and ascertain as accurately as possible the sound pressure distribution in the ear canal, we repeated and extended Wiener and Ross's measurements with somewhat modified equipment. While so doing, we paid particular attention to a careful determination of the points of measurement.

MATERIAL AND METHOD

Seven healthy randomly selected subjects (3 female and 4 male age 16-44 years) with clinically normal outer ear and normal hearing were used. The subject was positioned on an examination table in a soundproofed chamber

with acoustically treated walls and ceiling. The subject's head was placed sideways on a cork filled cushion (the left ear always towards the cushion) and fixed to the examination table by means of an adjustable belt. The right ear faced the sound source: an Altec 405E loudspeaker mounted on a square baffle (2 feet on each side) hanging from the ceiling, 110 cm above the entrance of the ear canal. A Brüel & Kjær Beat Frequency Oscillator Type 1022, served as the signal source. The frequency was swept from 20-20 000 Hz. The signals were amplified and delivered to the loudspeaker.

The sound pressure was measured by means of a Brüel & Kjær $\frac{1}{8}$ -inch condenser microphone connected to a 50 mm long tube of 1 mm outer diameter. Since we were only interested in sound pressure ratios at each frequency no probe tube corrections had to be introduced. Using a finely threaded screw at the microphone holder and a Zeiss surgical microscope the probe tube was advanced into the ear canal, and the sound pressure was measured at: (1) 1 mm above the tympanic membrane at the umbo, (2) 1 cm below the entrance of the ear canal, (3) the tip of the tragus, and (4) 1 cm above the tragus. The sound pressure was recorded on a Brüel & Kjær level recorder (2305A). The sound pressure at the above-mentioned locations in the outer ear and the length of the ear canal were measured 6 times on each subject.

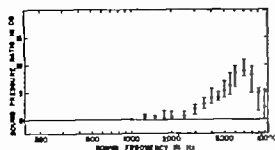


Fig. 1 Sound-pressure transformation between the eardrum and a point in the ear canal at 1 cm from the entrance. Circles indicate the medians; vertical range bars, the interquartile ranges.

RESULT

The mean length of the ear canals was 23.8 mm in the male and 22 mm in the female group. The overall mean of our population amounted to 23 mm. The results of the sound pressure measurements are shown in Figs. 1-4. They are expressed as ratios between the values obtained at the eardrum position and at the other positions.

Median sound pressure levels which are plotted in Fig. 2 by means of filled circles, do not generally follow the typical values. Because individual resonance frequencies vary along the frequency scale, the resulting median resonance frequencies vary along the frequency scale, the resulting median resonance peak should be somewhat broader than a typical peak. This is so mainly because

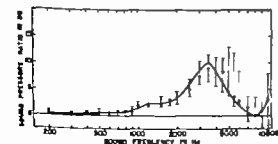


Fig. 2. Sound-pressure transformation between the eardrum and the entrance of the ear canal. Circles indicate the medians; vertical range bars, the interquartile ranges. The solid curve averages the data after normalization with respect to the frequency of the male maximum.

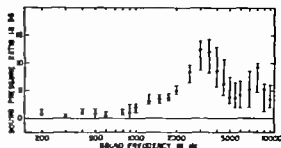


Fig. 3 Sound-pressure transformation between the eardrum and the tip of the tragus. Circles indicate the medians; range bars, the interquartile ranges.

peaks found in individual ears are not at exactly the same frequency. In order to investigate the size of the effect, all the individual data were normalized with respect to a median frequency of the peak, which was found to lie at 3.5 kHz. The solid curve in Fig. 2 approximates the normalized medians. It seems to be shifted somewhat towards lower frequencies relative to the non-normalized medians. However the width of the peak is practically unchanged. The correction introduced by the normalization may be regarded as negligible.

The sound pressure ratios between the eardrum and a position located 1 cm outside the tragus are plotted in Fig. 4. The first peak occurs at 3 kHz and the second, at 6 kHz. Both peaks are almost fused, so that a substantial enhancement of sound pressure takes place in the broad frequency range between 2 and 8 kHz. According to model experiments

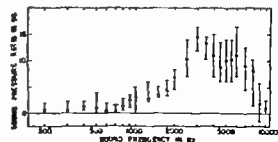


Fig. 4 Sound-pressure transformation between the eardrum and a point at 1 cm outside the tragus. Circles indicate the medians; range bars, the interquartile ranges.

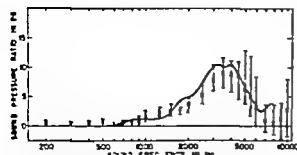


Fig. 5 Sound-pressure transformation between the ear drum and the entrance of the ear canal. Crosses indicate the means, range bars, the interquartile ranges. The solid line reproduces corresponding data of Wiener & Ross (1946).

by Shaw & Teranishi (1968), this effect is due to the quarter wave resonance of the concha.

DISCUSSION

Our data for the pressure transformation between the eardrum and the entrance of the ear canal deviate somewhat from those obtained by Wiener and Ross under similar conditions of the incident sound. The difference is illustrated in Fig. 5 where the solid curve shows Wiener & Ross's mean data, and the crosses, the means of our results. The main peak obtained by them is somewhat broader than in our investigation and is shifted somewhat towards lower frequencies. A secondary peak appears at 7.5 kHz. This pattern resembles the pattern found by us at the tip of the tragus (Fig. 3). We suspect that Wiener & Ross had the tip of their probe tube located between the entrance of the ear canal and the tragus rather than directly at the entrance. Our results are in close agreement with computations obtained with the help of an elec-

trical network analog (Zwislocki, 1965) and with data obtained by Teranishi & Shaw (1968) on a geometric model of the outer ear. The only appreciable difference occurs at the maximum and is most probably due to a difference between the acoustic impedance at the eardrum and the acoustic impedances terminating the model-ear canals.

Our data should be useful for practical purposes, such as the design of couplers for ear phone calibration, evaluation of ear protectors, and transfer of sound-pressure specifications from one point in the outer ear to another. From a fundamental point of view they demonstrate the contribution of the outer ear to the amazing sensitivity of the auditory system to sound. As is shown in Fig. 4 the outer ear enhances the sound pressure at the eardrum by between 1 and 9 kHz, a frequency range of 8 kHz. The maximum at 3 kHz amounts to about 15 dB.

REFERENCES

- Shaw E. A. G. & Teranishi, R. 1968 Sound pressure generated in an external-ear replica and real human ears by a nearby point source. *J Acoust Soc Amer* 44 240.
- Teranishi, R. & Shaw E. A. G. 1968 External-ear acoustic models with simple geometry. *J Acoust Soc Amer* 44 257.
- Wiener F. M. & Ross, D. A. 1946. The pressure distribution in the auditory canal in a progressive sound field. *J Acoust Soc Amer* 18 401.
- Zwislocki, J. J. 1965 Analysis of some auditory characteristics. *Handbook of mathematical psychology* (ed. Buse, Luce and Galanter). John Wiley and Sons, New York.

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COURSE OF AND SEQUELAE TO 248 PETROSAL FRACTURES

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Abstract The course of and sequelae to 26 transverse and 222 longitudinal fractures of the petrous portion are reported. Most patients were followed for a long time and seen 2-7 years after the trauma. Hearing loss of the conductive type has a very favourable prognosis, but for therapeutic and insurance reasons the patients should be followed.

A material of severe cranio-cerebral and other multiple traumas is difficult to follow and treat. This perhaps is the explanation why the literature contains only a few major series of petrosal fractures followed over a long period or after-examined. Owing to the good communication and collaboration between the specialities, we have been able to follow 248 petrosal fractures until the condition became normalized or stationary. The majority of patients were seen at follow-up 2-7 years after the injury.

The material comprises 230 patients admitted with fresh fractures from 1962-1968. In 18 the fracture was bilateral. The most common causes were automobile accidents and run-over accidents (42%), falling off bicycles, mopeds, or scooters (25%), fall or crash (28%). Children and adolescents predominated, only 41 patients being over 50 whereas 51 were 0-10 years. The petrosal fracture often co-existed with other cranio-cerebral injuries, such as cerebral contusion (25%), thecal fracture (27%), and concussion (35%).

Transverse fractures

Twenty-six patients (10%) had transverse fractures with destruction nystagmus, extinguished

vestibular function and anacusis. The two last-mentioned signs were also present at follow-up. Radiologically the fracture was demonstrable in 69% of the cases.

Longitudinal fractures

Longitudinal fractures were present in 222 cases and were demonstrated radiologically in 50%. According to the findings in the auditory canal and in the middle ear the longitudinal fractures were divided into cases with rupture of the drum (128), meatal fractures without rupture of the drum (38), and haematotympanum (51). In 41% of the cases of this latter group the fracture in the petrosal part could be demonstrated radiologically. In several cases it could be seen subcutaneously in the meatus or as a fracture of the theca coursing towards the petrous portion.

One week after the trauma the perforation had closed in all patients but 27. 3 weeks after in all but 7. Three months after the trauma the perforation had closed in all the patients, so that early myringoplasty is hardly indicated. Haematotympanum subsided in 3 weeks in practically all cases. The delayed closure of the perforation was due in 7 cases to major dry perforation, in 7 to persisting otorrhoea of cerebrospinal fluid which also subsided spontaneously in all cases, and in 13 to acute infection although 9 of them had received prophylactic penicillin. Prophylactic penicillin therapy probably does not reduce the incidence of infection and is in my opinion unnecessary.

Table 1 *Hearing in the 500-2 000 cps range primarily, 3-6 weeks after and 3 months to 7 years after the injury*

dB-Interval	Primarily	3-6 weeks after	3 mo.-7 years after
0-20	54	139	176
21-30	53	34	9
31-40	32	8	10
41-50	34	11	6
51-60	17	6	5
Over 60	11	5	6
Not examined	19	19	10
Total	222	222	222

113 patients were treated prophylactically with penicillin and 80 were not. 8% and 5% respectively developed acute infection which however did not cause major therapeutic problems.

Vestibular signs, such as spontaneous nystagmus, positional nystagmus, and reduced vestibular function were found initially in 22% also equally distributed on the various groups of fractures.

The primary hearing loss may often (in 24%) be within the normal range (0-20 dB Table 1). 3-6 weeks after it had returned to normal in 63%. The greater the primary conductive hearing loss the less the chance of rapid normalization and the greater the likelihood of dislocation or fracture of the ossicular chain. Some spontaneous improvement occurred in nearly all cases during the first 3 weeks, except in patients with perceptive hearing loss. If hearing of the conductive type was still poorer than 30 dB 6 weeks after the trauma, the cause was always ossicular dislocation or fracture. However spontaneous improvement may occur 6 weeks after the trauma, but it is slight and rare. The final hearing status 3 months to 7 years after the trauma was as follows: 80% had hearing within the normal range in the frequency group 500-2 000 cps, but in 20% the hearing of the conductive type was 5-10 dB poorer than in the non-damaged ear. In 21% there was slight perceptive hearing loss of 10-25 dB for the range 4 000-8 000 cps, and in

6% there was moderately severe hearing loss of 20-60 dB for the range 2 000-8 000 cps, but most marked for 4 000 cps. In 13% (28 cases) there was ossicular dislocation or fracture. Of these patients 10 had moderately severe to severe perceptive hearing loss of high tones and partially also of low tones. A total of 8 patients (3.5%) had perceptive hearing loss of low tones, not improved since the trauma.

Out of 28 patients with ossicular dislocation or fracture 16 underwent operation. 10 were not diagnosed until at follow-up, indicating that in spite of all the testing after the trauma failed in some cases. There were several types of dislocation, most often of the long process of the focus towards the handle of the malleus (5 cases) superiorly associated with fracture of the stapes (3 cases) or posteriorly (3 cases). As a rule interposition of the incus was performed. The results were good (Tos, 1971), giving an average hearing gain of 30 dB, average postoperative hearing 21 dB. 12 patients were not operated upon. Hearing improved spontaneously in 6, there occurring spontaneous fibrous connection between the stapes and incus or spontaneous myringo-stapedioplasty.

Peripheral facial palsy was observed in 44 patients. In 10 it occurred immediately after the trauma. In these cases the prognosis was poor, there being one year after the trauma—in spite of operation—still one total and 6 partial palsies. In 34 cases the palsy was tardive occurring 1-7 days after the trauma. In all these patients it had completely disappeared as early as 3 months after the trauma.

REFERENCES

- Tos, M. 1971. Prognosis of hearing loss in temporal bone fractures. *J Laryng* 83: 1147.

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FRACTURE OF TEMPORAL BONE, EARLY OR RETROSPECTIVE DIAGNOSIS, AND SURGICAL HEARING RECONSTRUCTION

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Abstract Among the victims of acute skull trauma, temporal bone fracture is found in 6-8%. An early diagnosis emerges from clinical inspection, X-ray exposure and is strongly supported by otoscopy. The otologist is concerned about the facial nerve immediately after the fracture and to detect signs of defects which call for decompression. The destiny of hearing is due either to cochlear damage or alterations in the ossicular mechanism. Conductive hearing loss possibly resulting from previous skull trauma may benefit from surgical intervention in the tympanic cavity.

All patients in Oslo who fall victim to head injury are admitted to the Department of Neurosurgery in the Municipal Hospital. During the years 1964-68 temporal bone fracture occurred in 8.4% of head traumas. During 1970-71 the rate was 6% and 120 patients were investigated as a material of interest within the field of traumatic otology.

Table I. Diagnostic results from X-ray examination (120 cases)

<i>Survey of skull on admission</i>	
Fracture of temporal bone demonstrated	90 (75%)
Fracture of temporal bone not demonstrated	30
<i>Special exposure of primary arc cases</i>	
Fracture of temporal bone demonstrated	14
Fracture of temporal bone not demonstrated	16
	104 (86.7%)

The diagnosis temporal bone fracture was identified clinically as mental bleeding in 76%. X-ray survey of the skull gave the diagnosis in 75%. Supplementary otoscopy demonstrated hemotympanum in those not having visible external bleeding. Guided by otoscopy which revealed hemotympanum special X-ray exposures of the temporal bone itself increased the total score of X-ray positive findings to 86%. In the remaining 14% the diagnosis had to be based upon the clinical examination (Table I).

Along with the fracture diagnosis the facial nerve function must be checked. Any paresis or paralysis registered within 12 hours after injury indicates a probable lesion of the nerve trunk, usually between the knee and the styloid.

Table II. Temporal bone fracture involving the facial nerve (120 cases)

Onset of palsy related to injury	Peripheral	Central
Within 12 hours	~	~
Between 12-48 hours	3	
After 2 days	4	
After 5 days	5	
	14 (11.7%)	
Restored facial function in	11	

1 nerve decompression performed. The patient died 2 months after injury. 1 patient died 4 days after injury. 1 patient died 3 days after injury. 1 patient alive with permanent palsy.

Table III *Hearing situation among surviving patients*

Included in scheme	120	
Deceased within 14 days	33	
Survivors		97
Subject with perceptive loss slight to severe	7 (7.2 %)	
destroyed cochlea	7 (7.2 %)	14 (14.4 %)
Subject to conductive loss transitory and restored to normal		78 (80.4 %)
qualified for hearing reconstruction		1 (1 %)
Not available for follow-up		4

foramen. Early appearance of nerve damage calls for decompression with opening of the facial canal. When paresis occurs after a latency of 24-48 hours the prognosis is good, even without operation (Table II).

During convalescence following the fracture one has to identify the nature of an initial hearing loss, whether conductive or perceptive. A fracture may cause damage to the ossicles. In some cases a subsequent exploration of the tympanic cavity discloses a disorder of the ossicular chain which can be restored (Table III).

This experience should encourage the oto-

Table IV *Repair of fracture: interval and X-ray fiction*

Case no.	Age	Interval between injury and op. (years)	Preoperative roentgenography	Observation period (years)
1	1	4/1	Fracture of pyramid	4/12
2	11	4/1	Fracture of squama	4/12
3	32	5/1	Fracture of squama	5
4	23	7/1	Fracture of squama	2
5	61	1	Fracture of squama	1
6	37	15	Negative	1
7	30	16	Negative	10, 1
8	22	16	Fracture of squama	-
9	65	33	Fracture of squama	14/1

Table V *Repair of fracture: findings and method*

Case no.	Age	Operational findings	Method	Thresholds and gain (dB)
1	12	Chain intact	Explor. only	65-65 - 0
2	11	Cranial fracture	Piston on plate	58-33 - 13
3	3	Luxation of stapes	Reduct.	50-10 - 40
4	23	Adhesions	Debrid.	38-23 - 15
5	61	Focus and stapes destroyed	Explor. only	58-50 - 5
6	37	Luxation of stapes	Reduct.	43-25 - 18
7	30	Long proc. defective	Incudo-staped. tubing	47-33 - 14
8	22	Long proc. missing	Interpos. of focus	47-8 - 39
9	65	Bridge fracture, mal. joint defect	Cloth pin synthesis	77-55 - 17

Table VI *Circumstances leading to temporal bone fracture (120 cases)*

Traffic accident	47 (39.2 %)
Violence or assault	14 (11.6 %)
Injuries other than above	59 (49.2 %)

logist to look for a possible connection between a conductive hearing loss and previous head injuries. The interval between a head trauma with persistent conductive loss and the retrospective diagnosis of temporal bone fracture varies from months to years. Special X-ray exposures and tomography of the temporal bone itself are a valuable aid to preoperative assessment (Table IV).

The case history, roentgenography and audiological measurements can justify exploratory measures. A great variety of ossicular defects are encountered and can be dealt with (Table V).

In large cities the majority of fractures are caused by traffic accidents. A large number arise from sports and industrial activity. A number of elderly people get injured in their homes (Table VI).

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ISOLATED FRACTURES OF THE STAPEDIAL ARCH

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Abstract The most frequent traumatic lesion in the middle ear is the luxation of the incus, which is seen in the three-quarters of all cases. The isolated fracture of the stapedial crura accounts for the majority of the remaining cases and contains interesting aetiological, diagnostic and technical operative problems.

MATERIAL

Out of 50 traumatic lesions of the ossicular chain we have seen 11 cases of that kind at the ENT-department in Aarhus. Seven out of these 11 patients are from 5 to 15 years old. Two resulting from transmeatal traumas, the rest from severe cranial traumas where 8 of these had been unconscious with bleeding from the ear. 8 of the patients were handicapped by a tinnitus which disappeared post-operatively.

Examinations and diagnosis

An X-ray examination including tomography of the temporal bone has proved worthless in this connection but was at least able to exclude a luxation of the incus. Only in 2 cases was a longitudinal fracture of the temporal bone demonstrated. Preserved reflex of the stapedius muscle was found in 4 cases and in 2 of these the reflex of the tensor muscle elicited *a.m.* Klockhoff was increased. As a sign of an increased looseness in the ossicular chain, ear drum system, we accepted this and therefore confirmed the suspicion of an isolated fracture of the stapes.

Operative findings

All cases showed a fracture of the stapedial arch close to the footplate as the only defect.

In 1 case, however, a small remnant of the stapes footplate was left on the anterior crus. In 3 cases the stapedial arch was displaced to the promontorium.

RESULT

Fig. 1 shows the average hearing improvement of each patient, illustrated by a column. Patients 1-4 had the sound transmission system re-established by means of the incus as a columella between the stapedial footplate and the ear drum. Nos. 5-7 had the function of the ossicular chain re-established by interposition of a polyethylene tube or like no. 8 an autogenous bone graft between the footplate and the incus. In nos. 9-11 a simple reposition of the fracture was performed with interposition of a small bone fragment in the fracture line, as in case no. 9.

In all cases the stapedial footplate was preserved. The postoperative hearing threshold is in all patients 30 dB or better and the average hearing gain for the whole series is about 27 dB.

DISCUSSION

The way in which the craniotraumatic stapes fracture arises is very complicated, and it might be seen together with a luxation or subluxation of the incus at least. The direction and course of the fracture lines in the skull together with the displacing forces at the moment of the trauma, the strength of the fixing ligaments, the joint capsules and ossicles must be pieces in that puzzle which

Table III *Hearing situation among surviving patients*

Included in scheme	120	
Deceased within 1-14 days	3	
Survivors		97
Subject to perceptive loss slight to severe destroyed cochlea	7 (7.2%)	14 (14.4%)
Subject to conductive loss transitory and restored to normal qualified for hearing reconstruction		78 (80.6%)
Not available for follow-up		1 (1%)
		4

foramen. Early appearance of nerve damage calls for decompression with opening of the facial canal. When paresis occurs after a latency of 24-48 hours the prognosis is good, even without operation (Table II).

During convalescence following the fracture one has to identify the nature of an initial hearing loss, whether conductive or perceptive. A fracture may cause damage to the ossicles. In some cases a subsequent exploration of the tympanic cavity discloses a disorder of the ossicular chain which can be restored (Table III).

This experience should encourage the oto-

Table IV *Repair of fracture. Interval and X-ray reduction*

Case no.	Age	Interval between injury and op. (years)	Preoperative roentgenography	Observation period (years)
1	1	4 1/2	Fracture of pyramid	~1 1/2
2	11	4 1/2	Fracture of squama	4 1/2
3	3	5 1/2	Fracture of squama	5
4	23	7 1/2	Fracture of squama	2
5	61	1	Fracture of squama	1
6	37	15	Negative	1
7	30	18	Negative	10 1/2
8	22	16	Fracture of squama	2
9	83	53	Fracture of squama	14 1/2

Table V *Repair of fracture findings and method*

Case no.	Age	Operational findings	Method	Threshold and gain (dB)
1	1	Chain intact	Explor. only	65-65 0
2	11	Cranial fracture	Piston on plate	58-33 +18
3	32	Luxation of stapes	Reduct.	50-10 40
4	23	Adhesions	Debrid.	38-28 10
5	61	Incus and stapes destroyed	Explor. only	58-50 5
6	37	Luxation of stapes	Reduct.	43-25 18
7	30	Long proc. defective	Incudo-staped. tubing	47-33 14
8	22	Long proc. missing	Interpos. of incus	47-8 39
9	65	Bridge fracture, malleol. joint defect.	Cloth pin synthesis	71-55 +17

Table VI *Circumstances leading to temporal bone fracture (120 cases)*

Traffic accident	47 (39.2%)
Violence or assault	14 (11.6%)
Injuries other than above	59 (49.2%)

logist to look for a possible connection between a conductive hearing loss and previous head injuries. The interval between a head trauma with persistent conductive loss and the retrospective diagnosis of temporal bone fracture varies from months to years. Special X-ray exposures and tomography of the temporal bone itself are a valuable aid to preoperative assessment (Table IV).

The case history, roentgenography and audiological measurements can justify exploratory measures. A great variety of ossicular defects are encountered and can be dealt with (Table V).

In large cities the majority of fractures are caused by traffic accidents. A large number arise from sports and industrial activity. A number of elderly people get injured in their homes (Table VI).

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ONE THOUSAND MAXILLO-FACIAL AND RELATED FRACTURES AT THE ENT-CLINIC IN GOTHENBURG

A Two-year Prospective Study

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Abstract. A prospective study of 1000 maxillo-facial and related fractures in 899 patients treated at the ENT-clinic in a city of 450 000 inhabitants during two years is reported with special reference to the frequency of different types of fractures and the etiology. Fractures caused by traffic accidents tended to be more serious but injuries from assaults were quantitatively predominant. Alcohol consumption was often involved in cases of fractures from assaults and falls. The "hidden" costs of alcohol abuse in the somatic care is pointed out.

In the town of Gothenburg with approximately 450 000 inhabitants there is only one ENT clinic. The maxillo-facial and related fractures seen at this clinic during the period July 1969 to June 1971 totalled 998 well-defined fractures which affected 899 patients. Often the care of the patients required the services of an odontologist and neurosurgeon, ophthalmologist and plastic surgeon, as additional consultants.

471 patients were treated as outpatients and 428 were hospitalized. 670 operations were performed on 622 patients. 277 patients were treated conservatively. The age distribution showed a majority of injured in the age group of 20-29 years. The sex distribution in the total material was 5 men to 1 woman. In the temporal fracture group the conditions were, men to women 10:1.

This study was supported by grants from AB Fylgta

Fracture distribution (Table I)

The number of nasal fractures was high, as expected (39%). The zygomatic-maxillary fracture group (22%) includes the important group of fractures which involve the orbits. The number of fractures of the mandible was high (20%) and this seems to imply the use of brute force in the material and makes the analysis of the causes specially interesting. The number of temporal fractures (7.5%) agrees well with the other material. The number of frontal sinus fractures seems low but the latter depends upon the fact that a relatively large number (17 cases) are again found in the combined fracture group. In the group of combined fractures are included 38 cases of the mid-face fractures (Le Fort I, II, III).

Table I *Fracture distribution*

	Number of	
	Patients	
	<i>n</i>	Fractures
Nasal fractures	350	39
Zygomatic-maxillary	196	22
Mandibular	183	20
Temporal	64	7.5
Frontal sinus	4	0.5
Combined	96	11
Total	899	998

Table II Causes of fractures

Fracture type	Cause				
	Traffic	Assault	Falls	Sport	Other
Nasal	37	160	66	60	27
Zygomatic maxillar	30	93	38	3	1
Mandibular	33	98	35	6	13
Temporal	24	13	5	3	3
Frontal sinus	-	1	0	1	0
Combined	46	33	14	1	-
Total	172 (19)	398 (44)	178 (20)	94 (11)	57 (6)
No. of patients 899					

Causes of fractures (Table II)

Assaults are a strongly dominant cause (44%) and the most common fracture is a nasal one. The high frequency of mandible fractures agrees well with the brute force trauma which is often given by kicking a person who has been knocked down.

Traffic injuries Fractures caused by traffic accidents accounted for 19% in the total material and were qualitatively more severe owing to the high frequency of combined fractures. Among the traffic injuries the number of patients with temporal fractures was highest (36 cases) when isolated fractures of the temporal bone and those in the combined fracture group are added together. Often the temporal fractures occurred in combination with skull base fractures and concussion.

Among traffic injuries 33 protected (car borne) road users were affected compared with 87 unprotected road users (not in cars). Drivers without safety belts were injured in 46 cases, while in 4 cases injury occurred to drivers with safety belts, but the latter were in any case only slight. None of the front seat passengers (19 cases) who were injured had worn safety belts. Even if the material in the viewpoint of safety belts and their importance is strictly limited, it is in agreement with the great traffic injury material which shows the value of safety belts *partly* by lowering the number of traffic injuries and *partly* by reducing the severity of

the injury (Bäckström, 1963). The same experience applies to the unprotected road user material (motorcyclists) of whom 12 did not use the crash helmet, while 7 did wear one. The helmet seems to have protected them against the severe injuries.

Alcohol and fractures

An attempt to judge the importance of alcohol in connection with injury has been carried out. "Under the influence of alcohol on the occasion of the injury" was the judgement on those who showed clear signs of intoxication, smelled of alcohol and/or admitted taking alcohol. The connection between drinking alcohol and fighting seems obvious. In the group of mandible fractures 61% of the patients were under the influence of alcohol, when the cause has been fighting. The proportion of inebriates was even higher in the case of assaults in the temporal (85%), the zygomatic maxillar (66%) and the combined fracture groups (66%). 80% of the patients with temporal fractures after falls were intoxicated.

The distribution of the days of the week showed that the frequency of fractures, as well as the frequency of fractures in connection with alcohol consumption, showed a concentration around the weekend and this was especially applicable to the relation between alcohol and assault.

The investigation of the connection between alcohol and the occurrence of fractures, is an example of the "hidden" costs of alcohol abuse within the somatic care and the retarding effect on the care of sick persons with other somatic diseases in a position, where the supply of medical care does not meet with the demand.

From the continuing follow-up of the patient material only a few viewpoints are mentioned: *Nasal fractures* in the majority of cases have been treated traditionally with reposition under local anaesthesia and plaster and the patients treated as outpatients. Corrective secondary

operations have only been necessary in a few cases.

Zygomatic-maxillary fractures have been treated according to Gillies (53%), or Gillies together with inter-osseous wiring (11%). Operation according to Caldwell-Luc with inter-osseous wiring was undertaken in 6% and Caldwell-Luc with antral packing in 11%. The relatively high frequency of operations according to Gillies with or without osteosuture speaks well for our good experience of this method.

Our attitude to *frontal sinus fractures* is active and 9 of 17 cases were operated upon with exploration in certain cases in combination with inter-osseous wiring or other form of fixation. Tomography agreed well with the findings at operation concerning the back wall of the frontal sinus. Neither in operated nor in non-operated cases have sequelae with liquor fistula or meningitis arisen.

The treatment of the *mandible fractures* may be seen in our second paper in this journal.

In conclusion we would like to stress the importance of training in this field of traumatology of the otolaryngologist and the odontologist. One of the arguments favoring this view is that these specialists are represented at the county hospital, where the fracture cases are mainly treated.

REFERENCES

- Blickström, C. G. 1963 Traffic injuries in South Sweden with special reference to medico-legal aspects of car occupants and value of safety belts. *Acta Chir Scand Suppl.* 308

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COMPLICATIONS AFTER JAW FRACTURES

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Abstract 260 consecutive cases of jaw fractures were seen during a two-year period. A follow-up 6 months after treatment covering 81 patients showed that the dental injuries are the most important sequelae of a jaw fracture. The complications noted at treatment had been fewer except for the disorders of the temporo-mandibular joint which had an increasing tendency. It was striking to note the great falling off percentage of suspected alcohol abusers, i.e. patients injured by assaults and falls.

Jaw fractures in "the Gothenburg fracture material July 1969 to June 1971" presented in our preceding paper together with jaw fractures from Gothenburg Children's Hospital and Mölndal County Hospital from the same period were treated in cooperation between otolaryngologists and odontologists. Patients with fractures of odontological interest were seen during the acute phase by a dentist at the Odontological Clinic, Gothenburg.

This report deals with the results obtained and special reference is given to the type and frequency of complications registered at the end of treatment and at follow-up 6 months later. The material comprised 260 patients. 212 cases (82%) were mandible fractures, 22 cases (8%) were mid-face fractures and 26 cases (10%) were combined mandible and mid-face fractures. With regard to the causes of the fractures the reader is referred to our preceding paper in this journal.

The treatment of the mandible fractures has fundamentally consisted of closed reduction and inter maxillary fixation. Inter-osseous wiring

has been used in some cases mainly to get control over edentulous jaw fragments. Usually this management has been combined with inter maxillary fixation. Instead of wiring plates has been used in a few cases.

Mid-face fractures have in general been treated with internal osseous fixation to the malar bone or to the lateral orbit-edge in addition to inter maxillary fixation. The period of immobilisation has been 2-9 weeks with an average of 4 weeks. The short fixation times have as a rule applied to condylar and mid-face fractures. Isolated condylar fracture was in most cases treated conservatively. Condylar fracture combined with other mandibular fracture were treated by fixation, the time being decided by the type of combined fracture symptoms and disturbances in the bite. Immediately after the immobilisation period jaw exercises were started.

Table I shows the type and frequency of complications at the end of the treatment. Teeth injuries were found in 67 cases (26%). A reduced ability to open the mouth—at opening the distance between the incisal edges of the frontal teeth was less than 40 millimetres—was noted in 49 cases (19%). Anaesthesia due to injury of the infraorbital or the inferior alveolar nerves was registered in 37 cases (14%). Disorders in the temporo-mandibular joint was observed in 9 cases (3%). Facial or jaw deformities or malocclusion were noted in a few cases.

So far 81 out of 260 patients have appeared

This study was supported by grants from AB Fylgia

Table I Complications at the conclusion of treatment

Type of fracture	No. of patients	Deformities of the face	Sensory disturb.	Temp.-mand. joint disorders	Restriction of opening	Mal-occlusion	Injuries of the teeth
Condyl.	60	1	1	4	9	1	8
Other mand.	101	3	16	4	11	1	24
Cond. + other mand.	51	—	6	—	20	2	17
Midface	22	—	6	—	1	1	13
Mandible + midface	26	3	8	1	8	1	5
Total	260	7	37	9	49	6	67

for the check-up 6 months after treatment. Presumably those with complications are over-represented in this group, but a striking feature is the large percentage of falling off among the patients with a history of alcohol abuse and/or patients injured by assaults and falls. Thirty-one of the 49 patients with restriction of opening were seen at follow-up and only 9 (11%) showed persisting restriction. Anaesthesia was found in 11% but the observation period was too short to evaluate whether the nerve injury was permanent or not. It may be noted that the anaesthesia in the mentals region did not show any quick regression. The frequency of disorders in the temporo-mandibular joint showed a tendency to increase already after this relatively short observation period.

In patients with complications after 6 months the follow-up period must of course be extended. Further cases of complications are also to be expected after this observation period, especially joint disorders. The dental injuries are of course permanent and often cause great psychological and also economical strain on the patients. In our experience quick reposition and fixation and administration of antibiotics, increase the chances for healing of injured teeth and alveolar processes. It is important not to neglect these injuries during the acute stage of the treatment of jaw fractures.

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CASES OF RECURRENT NERVE PARALYSIS IN GOTHENBURG FROM 1968-71

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Abstract. A study undertaken at the beginning of this century of the etiology of recurrent nerve paralysis was compared with a material from the present time. The comparison points to a changed pattern in the incidence of primary diseases.

Of all the works dealing with the etiology of recurrent nerve paralysis I have chosen to deal with the study by Paul Frenckner (1935) of 159 cases diagnosed at the ENT Clinic of Sabbatsberg Hospital, Stockholm, from 1903-1924. He also studied mortality for at least 10 years before publishing his results. The primary diseases behind the recurrent nerve paralysis in Frenckner's material are shown in Table I. As in most other investigations, the "unknown etiology" group was the largest. If cases diagnosed as "rheumatoid arthritis" are included, the group with an obscure etiology comprised 1/4 of the total number of cases. Gottle heads the list of primary diseases, including both benign and malignant thyroid enlargement. Mortality amounted no less than 60% during the period of observation more than half of the deaths occurring the first year after the discovery of the recurrent nerve paralysis.

The present study is based on 57 cases diagnosed as recurrent nerve paralysis at their first visit to the phoniatric department of the ENT Clinic, Sahlgrenska Hospital, Gothenburg, Sweden, from 1968-71. Most of the patients were referred to the phoniatric department from the ENT and other hospital clinics. In some of the cases, the purpose of the refer-

ral was evaluation of the indication for voice therapy. In other cases a subsequent follow-up by the phoniatrist was desired, even in the absence of voice disturbance. Postoperative cases following thyroid surgery were excluded from this material. The brief period of observation did not permit the tabulation of any relevant figures on mortality. A summary breakdown of the material into main diagnostic headings is presented in Table II. Paralysis unrelated to surgery occurred more than twice as often as in surgical cases. If the thyroidectomy cases had been included the relationship would have been the opposite. Thoracic surgery is now the predominant cause of paralysis in surgical cases, while neoplasms are responsible for the majority of the pareses due to non-surgical causes. As Table III shows, 5 of the cases of unknown etiology displayed other cranial nerve symptoms at the time of the onset of the recurrent nerve paralysis, symptoms such as external eye muscle paresis, facial nerve paresis and pharyngeal paresis. These symptoms were mostly reversible and of shorter duration than the recurrent nerve paralysis. "Virus disease" was used as the presumptive diagnosis in cases with an infectious course, since no serological or virus isolation tests were made. Table IV shows that melioidosis caused the largest number of cases of recurrent nerve damage among the different surgical procedures presented in the present material. Remarkably enough, this is also the only diagnostic one.

Table I. *Primary diseases in Frenchner's material from 1903-24*

	%
Unknown etiology	19.1
Gout	12.1
Pulm. tuberculosis	10.6
Aortic aneurysm	9.9
Oesophageal cancer	8.5
Syphilis	7.8
Rheum. arthritis	5.7
Organic heart failure	5.0
Tumour (pulm., pleural & mediastinal)	4.3
Mammary cancer metastasis	2.8

In conclusion if we look back on Frenchner's study from the first few decades of this century we should be aware that he excluded all postoperative cases which he regarded as being without prognostic relevance. Thus, his results can only be compared with the group of non-surgical cases in my material. Etiological sources such as gout, pulmonary tuberculosis, syphilis, unoperated aortic aneurysm and organic heart failure have become rare probably because of improved diagnostic and therapeutic methods. If the development of thoracic surgery, vascular surgery and neurosurgery permits successful treatment of many conditions which previously brought about recurrent nerve paralysis and death, it seems likely that the surgeon in our time plays a greater causative role in recurrent nerve paralysis. Publications in recent years (Hahn, 1970) even claim that the cuffed tube used during general anaesthesia may produce recurrent nerve paralysis as result

Table II. *Main diagnostic headings of the present material from 1968-71*

Diagnosis	No. of cases
<i>Non-surgical (40 cases)</i>	
Of unknown etiology	19
Tumour	10
Viral infection	6
Neurological disease	4
Trauma	1
<i>Surgical (17 cases)</i>	
Thoracic surgery (incl. mediastinoscopy)	11
Vascular surgery	3
Vagal neuroma surgery	3

Table III. *Etiology of the non-surgical cases*

<i>Unknown etiology (19 cases)</i>	
With other cranial nerve symptoms	5
Without other symptoms	14
<i>Tumours (10 cases)</i>	
Thyroid cancer	5
Lung cancer	2
Laryngeal cancer	1
Mammary cancer metastasis	1
Mb. Recklinghausen	1
<i>Viral infection (6 cases)</i>	
With influenza-like symptoms	6
<i>Neurological disease (3 cases)</i>	
Cerebral palsy	1
Polycaritis (Guillain-Barre)	1
Wallenberg syndrome	1
<i>Trauma (1 case)</i>	
Penetrating neck injury	1

Table IV. *Etiology of the surgical cases. Thyroid surgery cases excluded*

<i>Thoracic surgery (11 cases)</i>	
Mediastinoscopy	6
Aortic aneurysm	2
Botall's duct	1
Lung cancer	1
Benign pulmonary tumour	1
<i>Vascular surgery (3 cases)</i>	
Stenosis of com. carotid art.	1
Thrombosis of subclav. vein	1
Stenosis of subclav. art.	1
<i>Vagal neuroma surgery (3 cases)</i>	
Vagal neuroma	3

of local pressure damage. Parallels can easily be drawn with other medical fields in which prolonged patient survival has been accompanied by a higher incidence of various complications.

REFERENCES

- Frenchner P. 1935. The prognostic significance of paralysis of the recurrent laryngeal nerve. *Acta Otolaryng* (Stockh.) 34: 83.
 Hahn P. W. 1970. Vocal cord paralysis with endotracheal intubation. *Arch Otolaryng* (Chic.) 92: 226.
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ELECTROMYOGRAPHY IN MOTOR DISORDERS OF THE LARYNX

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Abstract EMG recordings have been performed from the cricothyroid and the vocal muscle in a series of patients with motor disorders of the larynx. Data obtained from 3 cases are reported. In one patient with idiopathic vocal cord paresis neurogenic lesions were found both in the vocal and in the cricothyroid muscle. In another patient subjected to thyroid surgery and in a third patient suffering from dysphonia bilateral lesions were demonstrated in the nerves to the cricothyroid muscles in both cases although there were no signs of abnormal mobility of the vocal cords at laryngoscopic examination. The results show that the EMG examination can disclose lesions in the motor nerves to the larynx which are not possible to reveal at clinical examination.

In a previous paper an electromyographic recording procedure was described which permitted identification of some 20 motor unit potentials from each normal vocal muscle (Knutsson et al. 1969). In that investigation 469 motor unit potentials from 18 normal muscles were identified, and the data collected could be used as a basis for prognostic evaluation in patients with a traumatic injury to the recurrent laryngeal nerve (Haglund et al., 1970) and also for an analysis of idiopathic vocal cord paresis (Haglund et al., 1972).

The results obtained in a recent investigation on the normal electromyogram in the human cricothyroid muscle (Haglund, 1973) provide further opportunities to reveal the existence of lesions of the motor nerves to the larynx which cannot be disclosed by e.g. indirect laryngoscopy, stroboscopy and X-ray examination. The present paper will describe the results obtained by electromyographic ex-

aminations of the cricothyroid muscle and the vocal muscle in some patients with motor disturbances of the larynx.

Case Reports

Case 1 was a 39-year-old man suffering for 7 weeks from a hoarseness with sudden onset. Indirect laryngoscopy showed that the left vocal cord was immobile in a paramedian position, but extensive clinical investigations failed to reveal any underlying cause and the paresis was thus classified as an idiopathic vocal cord paresis. In Fig. 1 the hatched columns of the histograms show the distribution of duration and amplitude of 13 motor unit potentials from the left cricothyroid muscle; for comparison are included corresponding data from 338 motor unit potentials identified from 17 muscles in 13 normal subjects. In this case several of the potentials were of normal amplitude and duration, showing that a considerable proportion of the motor axons in the external branch of the superior laryngeal nerve were conducting nerve impulses. However in addition there was an increased incidence of potentials of a duration less than 5 ms and, though not shown in the histogram, these potentials were of an amplitude below 150 mV thus indicating that they were fibrillation potentials. Thus many of the motor fibres were denervated. Electromyographic recordings from the left vocal muscle on the same occasion disclosed electrical silence indicating a total paresis of this muscle (cf Haglund et al., 1971).

Case 2 was a 34-year-old man who had been subjected to bilateral resection of the thyroid gland because of atoxic adenomas. Since that time his voice was easily fatigued and he was unable to speak or sing at a high pitch. Indirect laryngoscopy immediately after the operation, as well as at the time of EMG recording 36 months later, showed that the vocal cords had normal motility though the position of the right vocal cord was slightly lower than the left. As appears from the histogram in Fig. 2 the left cricothyroid muscle had an increased incidence of potentials of a duration less than 5 ms. These pot-

M crico thyroideus

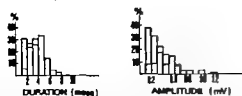


Fig. 1 Duration and amplitude distribution of motor unit potentials recorded from cricothyroid muscle in Case 1 (■) as compared with corresponding data from the normal material (□).

M crico thyroideus

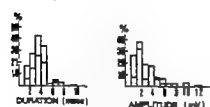
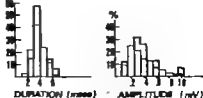


Fig. 2 Duration and amplitude distribution of motor unit potentials recorded from cricothyroid muscle in Case 2 (■) as compared with corresponding data from the normal material (□).

tial's were of low amplitude, indicating denervation. In addition, out of the 35 potentials recorded 24% were polyphasic as compared with only 0-4% in the normal muscle (Haglund, 1972). These potentials were of high mean duration, indicating that re-innervation had taken place in part of the previously denervated muscle fibres. EMG recording from the right cricothyroid muscle showed signs of increased insertion activity and only four potentials were found, two of which had the characteristics of fibrillation potentials, thus indicating that a lesion was present also in the right external branch of the superior laryngeal nerve. Thus, while laryngoscopic examination in this case failed to give any conclusive information, EMG recording was a valuable adjunct in analysing the etiology of the voice disturbance.

Case 3 was a 17-year-old girl suffering from voice disturbances since childhood. The most prominent abnormality was a monotonous voice and inability to raise the pitch of the voice. The vocal cords had normal motility and position, and the patient was treated under the diagnosis of dysphonia. Fig. 3 shows the duration and amplitude distribution of the action potentials recorded from the right vocal and cricothyroid muscles. The potentials recorded from the right vocal muscle are of normal duration and amplitude and the interference pattern is complete. In the right cricothyroid muscle, however the mean duration of the potentials is increased and there is an increased incidence of potentials of long dura-

M. vocales



M. crico thyroideus

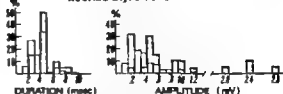


Fig. 3 Duration and amplitude distribution of motor unit potentials recorded from vocal muscle (above) and cricothyroid muscle (below) in Case 3 (■) as compared with corresponding data from the normal material (□).

tion (range 6-9 ms) and such of high amplitude (range 900-2 800 microvolt). In addition, the majority of these potentials were polyphasic, thus indicating that re-innervation had taken place. The muscles on the left side showed a similar pattern, i.e. a normal pattern in the vocal muscle but in the cricothyroid muscle there were electromyographical signs of a lesion in part of the motor axons to the muscle.

The data presented show that EMG recordings from the laryngeal muscles may be of great value in voice disturbances of unknown origin.

REFERENCES

- Haglund, S. 1973. The normal electromyogram in human cricothyroid muscle, *Acta Otolaryng (Stockh.)* in press.
- Haglund, S., Kuntzson, E., Mårtensson, A. & Mårtensson, B. 1970. Electromyography in vocal cord pareses, *Acta Otolaryng (Stockh.)*, Suppl. 263: 85.
- Haglund, S., Kuntzson, E. & Mårtensson, A. 1972. An electromyographic analysis of idiopathic vocal cord paresis, *Acta Otolaryng (Stockh.)* 74: 265.
- Kuntzson, E., Mårtensson, A. & Mårtensson, B. 1969. The normal electromyogram in human vocal muscles, *Acta Otolaryng (Stockh.)* 68: 256.

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ENDEMIC RECURRENT LARYNGEAL NERVE PARESIS

Correlation between EMG and Stroboscopic Findings

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Abstract. Sixteen cases of recurrent laryngeal nerve paresis are presented, 8 bilateral and 8 unilateral. 13 of the 4 paretic vocal cords also had paresis of the laryng. vap. Some patients had paresis of the face, tongue, sternum or diaphragm. Only 3 vocal cords in 5 patients regained normal mobility during the observation period, one year and a half. EMG was correlated with stroboscopic findings and with the position of the vocal cords. Considerable reinnervation and normal vibratory mobility was found in 1 vocal cord within 10 months, but of these only abducted and adducted normally after more than 1 year. Our conclusions were: 1) Glottic wave depends on active tone, i.e. innervation of the muscle in the actual vocal cord but does not necessarily imply functioning innervation. 2) The position of the paretic vocal cord depends, at least in part, on the degree of paresis.

indicating affection of other nerves. Most of them had difficulty in swallowing some of them so severe that they rapidly lost weight. Occasional paresis of the facial, vagus, hypoglossus and phrenic nerves were also found. In addition, several of the recurrent laryngeal nerve paresis cases seemed to be complicated by paresis also of the superior laryngeal nerves.

The diagnosis superior laryngeal nerve paresis, however, could not be made clinically in some of the cases, at least not from the supposition that if such a paresis was combined with a homo-lateral recurrent laryngeal nerve paresis the vocal cord should be in the intermediate position. In reality it was in such a case the quality of the voice and not the local findings, which aroused the suspicion, later to be verified by the use of EMG.

Larynx was inspected at rather short intervals using stroboscopic light. In 17 patients, 4 with bilateral and 8 with unilateral paresis of the recurrent laryngeal nerves, EMG was made from the vocal muscles and the cricothyroid muscles on both sides. Concentric EMG needles were used and put in place percutaneously. EMG was made on two occasions, about 5 months and about 10 months after the occurrence of the paresis.

To judge from the first EMG investigations 13 of the cricothyroid muscles were more or less paretic, 2 bilateral and 11 unilateral. Cor-

During the winter 1969-70 an increase in the incidence of recurrent laryngeal nerve paresis appeared in several parts of Europe (Bauer 1971, Hefter & Bildstein 1970, Wirth & Leyboldt 1970). The patients were said to have been ill a few weeks previously with the so-called Hongkong flu. After dubious cases had been sorted out 9 men and 7 women were left in our material, none of them younger than 30 years, with altogether 8 bilaterally and 8 unilaterally immobilized vocal cords. In spite of thorough investigations it has not been possible to ascertain the genesis, but the viral infection has been supposed to be the origin although the connection is somewhat uncertain.

Fifteen of the patients also had symptoms

siderable reinnervation could be registered at the second EMG in all investigated cricothyroid muscles.

Out of the 24 paretic vocal cords normal or almost normal mobility could be found in only 5 after the complete observation period, a year and a half but none of these until after the first half year. This is considerably later than is usually the case in paresis of the recurrent laryngeal nerve.

At the second EMG investigation a good reinnervation could be registered in 12 of the still paretic vocal cords and at the same time good or even excellent glottic wave could be seen in stroboscopic light. Of these 12, however only 2 abducted and adducted normally after further more than 2 months, i.e. more than 1 year after the patients fell ill. In not a single one of the remaining immobilized vocal cords in this group with good reinnervation and good glottic wave was normal mobility found during the rest of the observation time.

As far as other symptoms are concerned it should be mentioned that all patients except one swallowed normally after usually less than half a year that a paresis of the facial nerve disappeared after less than a month, while a deviation of the velum in 1 patient and the deviation of the tongue in another patient remained unchanged during the whole period.

If EMG findings and stroboscopic findings are correlated it can be noticed that the presence of reinnervation in the vocal muscle corresponds very nicely to the occurrence of glottic wave in the vocal cord. As such a glottic wave can always best be seen at so-called chest register when the vocal muscle is active but not at so-called falsetto when the muscle is inactive it seems reasonable to suppose that the glottic wave depends on active tonus, in other words on the innervation of the muscle in the actual vocal cord (Fex, 1970). It should be stressed that such an innervation of one single muscle does not necessarily imply func-

tional innervation in all the laryngeal muscles, which of course is necessary if one expects coordinated vocal cord function. In order to know the innervation of the different laryngeal muscles it is necessary to examine every single muscle with EMG. To form a general opinion and especially one about the state of innervation in the vocal muscle at phonation, however it is recommended to use the stroboscopic examination which is easy to perform and easy to undergo. In other words, it is clinically practical.

The investigation also shows that just as partial paresis can be found in other parts of the body it can be found in the laryngeal muscles. This would appear to be completely obvious but has generally been surprisingly disregarded. In reality one has to take into account the occurrence of partial paresis as well as nonfunctional reinnervation. This is no doubt of importance for the position of the immobilized vocal cord. Likewise a moderate increase of innervation in a paretic cricothyroid muscle may suffice to explain a change in position from intermediate to paramediate in a paretic vocal cord.

REFERENCES

- Bauer H. 1971 Klinische Besonderheiten der Vagus-Neuritis nach Grippeepidemie. *Arch Klin Exp Ohr Nas Kehlkopfheilk* 199 731.
 Fex, S. 1970. Judging the movements of vocal cords in laryngeal paralysis. *Acta Otolaryng* (Stockh.), Suppl. 263 82.
 Hefter E. & Bülstein, P. 1970. Endemisches Auftreten von Rekurrensparesen im Winter 1969/70. *Z Laryng Rhinol Otol* 49 787.
 Wirth, G. & Leyboldt, R. 1970. Gehäufes Auftreten von Stimmbandlähmungen während der Grippeepidemie im Winter 1969/70. *Z Laryng Rhinol Otol* 49 777.

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VOCAL CORD PALSIES FOLLOWING SURGERY FOR BENIGN NON TOXIC GOITRE

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Abstract The frequency of palsies of the recurrent nerve in a series of 271 patients undergoing surgery for nontoxic benign goitre is analysed in respect to the number of operated sides.

Meurman (1951) and Evoy (1961) have stated that the incidence of vocal cord palsies after thyroid surgery varies between 0.3% and 13.2% with primary operations and that it is much higher in reoperated cases. The reason for the variability is that the series are mixed regarding preoperative assessment, pathology and method of operation.

METHOD AND MATERIAL

Since 1963 at the E.N.T. clinic in Örebro we have used uniform methods for preoperative assessment, operative technique and postoperative follow up. All patients subjected to thyroid surgery from 1964 to 1970 are included in this series except for cases of thyroid cancer or toxic goitre. The vocal cord function of all the patients was checked twice preoperatively. Four types of operation were used.

1 Enucleation of adenoma

In these cases, *n. recurrens* was not identified and the thyroid arteries were not divided. This method has only been used in case of superficial adenomata and when the risk of injuring *n. recurrens* has been judged as non-existent.

2 Resection

The *n. recurrens* was identified, the anterior thyroid art. was divided and the *n. recurrens* was resected. Only when a subtotal lobectomy was this r

3 Subtotal lobectomy

The *n. recurrens* was identified, both the superior and inferior thyroid arteries divided and a thyroid remnant was left.

4 Total lobectomy

The *n. recurrens* was identified and the arteries were divided as in group 3. The surgery was performed leaving no macroscopic evidence of thyroid tissue.

Postoperatively the patients have been followed up for 1 to 8 years. Indirect laryngoscopy was performed at least 3 times in the first year and yearly thereafter. The series consists of 283 patients (235 women and 48 men) of which 264 had adenomatous nontoxic goitre or solitary adenomata and 19 had thyreoiditis or adenomatous non toxic goitre with a proven histology of thyreoiditis. The second group were usually subjected to surgery because of suspected malignancy.

Nine patients with preoperative palsies have been excluded from this series. One case had a large goitre and had emergency surgery because of respiratory obstruction. On preoperative examination the vocal cords were found in almost paramedian position but adduction was present. At operation the right *n. recurrens* was bluish and swollen which can be taken as a sign of degeneration. Postoperatively there was a right irreversible *n. recurrens* palsy. Three patients with preoperative palsy eventually showed normal vocal cord function postoperatively. That vocal cord palsy can be seen in non-malignant goitre is re-

DISCUSSION

The total number of palsies following primary thyroid surgery in 261 patients was 17 (6.5%) of which 3 (1.1%) were irreversible. After 46 enucleations and 68 resections there were no irreversible palsies. In order to see the result of exposing the recurrent nerve one can compare the 144 subtotal lobectomies and the 92 total lobectomies with the corresponding groups of non-toxic goitre in Gisselsson's (1950) report. In his series there were 52 total lobectomies and 104 subtotal thyroidectomies, making a total of 260 (52 + (2 × 104)) sides with 14 (5.4%) palsies of which 6 (2.3%) were irreversible. In Gisselsson's cases the recurrent nerve was not exposed.

In the Örebro material there were 236 sides (144 + 92) in the subtotal and total lobectomy groups resulting in 19 (8%) palsies of which 3 (1.3%) were irreversible. In comparing these series it is found that a higher total number of nerve injuries results when the nerve is exposed but few irreversible palsies. Lahey & Hoover (1938) reported that in 3 000 cases they reduced the number of postoperative palsies to 0.3% by exposing the nerve. In the re-operated group there were 3 irreversible palsies in 13 patients (17 sides) which illustrates the far greater risk of nerve injury in these cases.

REFERENCES

- Evoy M 1961 Paralysis of vocal cords after thyroidectomy *Amer J Surg* 102 73
 Gisselsson, L. 1950. Laryngeal paralysis following thyroidectomy *Acta Chir Scand* 99 1950
 Hall-Allen, R. 1967 Laryngeal nerve paralysis and benign thyroid disease. *Arch Otolaryng (Chic.)* 85 335
 Lahey F & Hoover W 1938 Injuries to the recurrent laryngeal nerve in thyroid operations. *Amer J Surg* 545 1938
 Mourman, O H 1951 Vocal cord paralysis following thyroid surgery: A study of 104 cases. *Acta Chir Scand* 101 1951

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Table 1 Number of operated sides and palsies (261 patients having primary thyroid surgery)

Type of operation	No. of sides	Total no. palsies Right/Left	Irreversible palsies Right/Left
Enucleation of isthmus adenoma	8	0/0	0/0
Enucleation of adenoma 20/18	38	0/0	0/0
Resection 32/36	68	1/0 (1.5)	0/0
Subtotal lobectomy 61/83	144	6/6 (8.3)	2/0 (1.4)
Total lobectomy 48/44	92	2/2 (4.4)	1/0 (1.1)

RESULT

In order to judge the risk of dissecting the n. recurrens the number of sides operated have been used in this account. Therefore one patient can be accounted for twice if he had bilateral surgery. Eight cases with only isthmus adenoma are not referred to as any side.

The table shows the number and distribution of operated sides and how the palsies are distributed between the different types of operation. After 46 enucleations and 68 resected sides there were no irreversible nerve palsies. Subtotal lobectomy was performed on 144 sides and there were 12 (8.3%) palsies of which 2 (1.4%) were irreversible, and total lobectomy was performed on 92 sides and there were 4 (4.4%) palsies of which 1 (1.1%) was irreversible.

No case of bilateral palsy was seen. The 3 irreversible palsies were all on the right and none were found in the thyroiditis group.

Thirteen cases having previous thyroid surgery developed 3 irreversible palsies in 17 operated sides.

EXAMINATION OF VOICE FUNCTION OF PATIENTS WITH PARALYSIS OF THE RECURRENT NERVE

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Abstract 96 patients were examined when the diagnosis of paralysis was made, with indirect laryngoscopy determination of pitch and voice range, phonation time, respiration, stroboscopy and, in addition, oscillography was carried out. Speech therapy was carried out for 64 patients and follow-ups were made with electroglottography and the above-mentioned parameters. The methods and preliminary results are discussed.

During recent years we have seen between 20 and 25 patients with paralysis of one or both recurrent nerves per year. The patients have been examined in the ENT department, and if possible also by a speech therapist. We give here some information of the results of 96 patients, 70 women and 26 men, examined during the last 4 years.

Fifty-two patients had their recurrent nerve paralysis after surgery for benign thyroid and parathyroid disorders. 11 patients had no other symptoms. 10 patients had malignant tumours, 7 cases have been diagnosed after influenza, 6 patients had intracranial disease and 4 patients had cardio-vascular disease. 1 patient had had a trauma of the thorax, 1 patient had had thoracic sympathectomy performed bilaterally and 1 patient had brachyoesophagus and a diverticulum of the esophagus. In 1 patient paralysis probably was due to compression from an enlarged thyroid gland, in 1 patient due to direct trauma and in 1 patient probably due to compression from a pulsating mediastinal tumour.

Nine patients had paralysis of both recurrent

nerves, 30 patients had right sided paralysis and 57 patients left-sided paralysis. 73 of the 96 patients had the paralyzed vocal cord in paramedian position.

Phonation time was, on average 8 seconds, the respiration mostly costal. At stroboscopic examination of 53 patients, among other parameters the amplitude was examined. 21 had an increased, 25 a reduced amplitude. 7 had normal amplitude. All had irregular vibration pattern. The voice range was much reduced, the pitch also reduced, on average.

The auditory examination and control of treatment by the speech therapist was felt insufficient and an oscillographic description of the tape recordings that were routinely taken was attempted. Among others, Smith (1961) and Lauritzen & Frøkjær Jensen (1970) have tried to evaluate the effect of speech therapy and have found that the sound spectrum over basic pitch frequency is intensified and that noise on voices with paralysis disappears. Among others Kolke (1968) and v. Leden & Kolke (1970) have shown that the vibration pattern of the vocal cords in patients with paralysis of the recurrent nerve is irregular.

We have tried to evaluate whether these parameters, 1) regular/irregular curve, 2) overtones, and 3) evaluation of noise could give us further information that would be useful in our practical work. This evaluation has till now been done by ourselves without mathe-

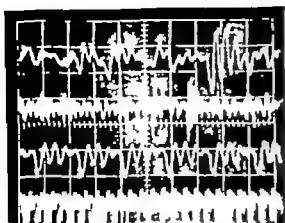


Fig. 1 Oscillometric curves. The upper and the third curve show 5 vibrations of a male with unilateral recurrent nerve paralysis before and after voice therapy. The second and fourth curves show the same voice at higher oscilloscopic speed.

mathematical calculation. We think that insufficient practical work has been done hitherto, to determine which parameters are most valuable for mathematical calculation.

In Fig. 1 are seen 2 oscillometric recordings, before and after voice treatment. The upper shows an irregular curve lack of overtones and noise. The lower shows regularity in vibration pattern, overtones and a smooth curve without noise.

At a follow-up examination of 47 patients, performed on an average 2 years after the diagnosis was made, 20 patients had persisting unilateral paralysis, 18 had no paralysis, 8 had some movement of the previously paralysed

vocal cord and 1 had a persisting bilateral paralysis.

The 20 patients with unilateral paralysis had an average phonation time of 11 seconds, pitch was normalised (215 Hz for women, 115 Hz for men) Voice range was, on average, 19 semitones. The stroboscopic examination was very uncertain, there was some movement of the paralyzed cord in all cases except 3.

Apart from the oscillographic examination, electroglottography was also carried out, where we tried to distinguish between 1) long/short closure, 2) identical/nonidentical vibration pattern and 3) even/uneven course.

Our electroglottographic results were not so clear as we had expected. This may be due to our evaluation method, which has to be refined in the direction recently described by Reinsch & Gobach (1972). We found a rather short closing phase at 10 of 18 patients without paralysis, in 1 of these no oscillography was done and in 9 of 20 patients with unilateral paralysis. This corresponded to the stroboscopy which showed poor voice function in many of those patients without paralysis. So there was good harmony between the stroboscopy and the electroglottography. The oscillography was better when it came to the description of differences between patients with and without paralysis, possibly because it could be called a more "summarizing" examination.

Sixty four patients had voice therapy and were all treated after Smith's accent method.

Table I Oscillometry

	At time of diagnosis (-16)	After voice therapy when no follow-up	At follow-up persisting paralysis	At follow-up no paralysis
Number of patients	80	32	20	17
Regular vibration pattern	21	22	13	13
Irregular vibration pattern	59	10	7	4
Many overtones	7	14	7	12
Some overtones	14	13	7	4
Lack of overtones	59	5	6	1
- none	35	24	19	15
noise	45	8	1	2

On average the patients had 15 treatments. We think that the best results are probably obtained after more than 10 treatments of one half-hour each.

We have tried to find some practically usable and simple methods for measuring voice disorders, not only paralysis of the recurrent nerve. At the same time we feel that we have got far better evaluation possibilities for voice therapy. Our present examination program now comprises indirect laryngoscopy examination of pitch and voice range, phonation time, respiration, stroboscopy, tape recordings, oscillography and electroglottographic examination.

EQUIPMENT

Stroboscope, Brüel og Kjær type 4910, with frequency analyzer type 2107 and condenser microphone type 4131.

Storage oscilloscope, Tektronix, type 564B, model 121N with a polaroid camera.

Tape recordings were performed on an Akai X 360 recorder frequency range 30-20 000 Hz with condenser microphone CM 15 and transferred to the

oscilloscope with an UHER 4000 report L, frequency range 40-20 000 Hz.

The electroglottograph was built by B. Frøkjær Jensen after the principle of Fabre.

REFERENCES

- Kolke, Y. 1968. Vowel amplitude modulations in patients with laryngeal diseases. *J. Acoust. Soc. Amer.* 43 839.
- Lauritzen, L. & Frøkjær Jensen, B. 1970. Comparative phonetic-acoustic analysis before and after speech therapy of voices suffering from recurrent paralysis. *ARIPUC 4* Institute of Phonetics, Copenhagen.
- von Leden, H. & Kolke, Y. 1970. Detection of laryngeal disease by computer technique. *Acta Otolaryng.* (Stockh.) 91 3.
- Reinach, M. & Gotsch, H. 1972. Zur quantitativen Auswertung elektroglossographischer Kurven bei Normalpersonen. *Folia Phoniat.* 24 1.
- Smith, S. 1961. On artificial voice production. *Proc. IV Int. Congr. Phonetic Science Helsinki.*

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VOCAL CORD PARALYSIS TREATED WITH TEFLON IMPLANTATION

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Abstract Nine cases of unilateral vocal cord paralysis with severe hoarseness combined with atrophy of the cord and asymmetry of the larynx were treated with implantation of Teflon in the paralytic vocal cord by Arnold's method. The voice quality was studied and judged by a listener-jury. After implantation the range of the voice was widened, the maximum phonation-time was remarkably prolonged and the quality of the voice improved significantly.

Training of the voice in cases of unilateral vocal cord paralysis most frequently gives a good result. The mobility of the healthy cord improves, resulting in closure of the ligamentous part of the glottis and the voice of the patient becomes clear. Sometimes, however, no result can be achieved by training-treatment. This applies to cases where an atrophy of the vocal muscle in the paralytic vocal cord has occurred. If the edges of the cords do not meet on the same level the dysphonia becomes even more difficult to control. In these cases the glottis is not only quite inadequate as a tone-generator but coughing and creating abdominal tension also become more difficult.

Brünings's idea (1911) to inject paraffin into the paralysed vocal cord was in principle excellent, but paraffin had to be replaced with something else which was indifferent and stable. Such a material has been found in Teflon, which is a polymer of tetrafluoroethylene. Pulverized Teflon in 50% glycerol suspension gives a paste which is easy to inject into the vocal cord. For this purpose a special injection-pistol developed by Brünings

is used. Arnold in particular has published studies on this topic during the years 1955-64.

The implantations in the present study have been done according to Arnold's technique (1967). Tomography of the larynx is very helpful when planning the procedure. In the ENT Department of Helsinki University Central Hospital these operations were performed under general anesthesia using Kleinsasser's microlaryngoscopy equipment.

The material comprised 9 extremely hoarse patients. In 5 cases the etiology of the paralysis was a struma-operation. The remaining 4 cases were 2 lesions of the vagus nerve caused by removal of tumour, one idiopathic paralysis of the vocal cord, and one patient was injured during the war by a shell fragment in the neck.

Larynx-tomograms were taken during breathing and intonation before and after Teflon implantation, stroboscopy was done and the maximum range of phonation (MFP) was measured. The pitch of the spoken voice and the physiologic tone-range were also measured. Tape recordings were made, and these samples were judged by a listener-jury who classified them in five categories according to the quality of the voice. Taking the mean of the judgements, a voice-quality index was obtained. In the statistical handling of the material a paired *T*-test was used (the paired *T*-test of difference, Ipsen & Feigl, 1970).

Common to all the patients before im-

On average, the patients had 15 treatments. We think that the best results are probably obtained after more than 10 treatments of one half-hour each.

We have tried to find some practically usable and simple methods for measuring voice disorders, not only paralysis of the recurrent nerve. At the same time we feel that we have got far better evaluation possibilities for voice therapy. Our present examination program now comprises indirect laryngoscopy, examination of pitch and voice range, phonation time, respiration, stroboscopy, tape recordings, oscillography and electroglottographic examination.

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REFERENCES

- Kolke, Y. 1968. Vowel amplitude modulations in patients with laryngeal diseases. *J. Acoust. Soc. Amer.* 43: 839.
- Lauritzen, L. & Frøkjær Jensen, B. 1970. Comparative phonetic-acoustic analysis before and after speech therapy of voices suffering from recurrent paralysis. *ARIPUC 4* Institute of Phonetics, Copenhagen.
- von Leden, H. & Kolke, Y. 1970. Detection of laryngeal disease by computer technique. *Acta Otolaryng.* (Stockh.) 91: 3.
- Reinach, M. & Gobsch, H. 1972. Zur quantitativen Auswertung elektroglottographischer Kurven bei Normalpersonen. *Folia Phoniat.* 24: 1.
- Smith, S. 1961. On artificial voice production. *Proc. IV Int. Congr. Phonetic Science* Helsinki.

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WHAT IS PROLONGED INTUBATION?

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Abstract The risk of damage to the upper airways by nasotracheal intubation is influenced not only by its duration, but also by a number of other factors. Among 29 patients treated by nasotracheal intubation, as a rule for 2-3 days, none showed laryngo-tracheal damage after extubation. Among 20 patients intubated for a longer period (5-54 days) numerous cases of damage to the respiratory tract were found. Prolonged intubation should be taken to mean intubation lasting longer than 6-7 days. With prolonged intubation the risk of severe damage to the larynx and trachea is so great that the possibility of replacing it by tracheostomy should be seriously contemplated.

Through the past 20 years oro- and naso-tracheal intubation has been gaining ever increasing ground in the treatment of pulmonary insufficiency. In Scandinavia this development was triggered by the polio epidemic of 1952 in Copenhagen, when tracheal intubation was performed through tracheostomy (Lassen, 1956). The hazards involved by an oro-naso-tracheal tube especially to the larynx and subglottis, were soon realized, and it was generally considered that the tube had to be removed within 12-24 hours (Lewy & Sjöbert, 1951; Dam & Zwergius, 1952). As recently as 1962 Bergström recommended removing nasotracheal tubes at the end of 24 hours.

Since that time, opinions have become more liberal, and it was considered justified to continue the intubation for 2 or 3 days (Fearon et al., 1966; Hatch, 1968; Hedden et al., 1969). In Denmark too nasotracheal intubation of an average duration of 2-3 days was reported in a series of patients without deleterious effects (Hansen & Jørgensen, 1968; Triff & Tox, 1969).

In the literature "prolonged" intubation is often mentioned without any explanation of what "prolonged" is taken to mean. Many variables play a role in damaging the respiratory tract. The general condition of the patient, the state of consciousness, premorbid respiratory infections and age. The material of the tube, the cuff pressure, and the frequency of changing the tube are also contributory pathogenetic factors.

In the present study it was endeavoured to ascertain for how long nasopharyngeal intubation can be maintained without a risk of clinical damage to the larynx and trachea.

Present investigations

Twenty-nine cases of acute epiglottitis in children were treated with nasotracheal intubation and antibiotics.

The majority were intubated for 2 or 3 days, and after extubation no patient of the entire group exhibited lesions of the larynx or trachea that required treatment. There was merely some hoarseness or stridor for a few days after extubation.

In another series comprising patients with respiratory insufficiency often of complex genesis, the nasopharyngeal intubation was of longer duration.

The primary disease that provided the indication for intubation was pulmonary in 9 whereas 8 patients had cerebral contusion. 3 patients were intubated for various reasons. The patients of this group were in an extremely poor condition, 17 were primarily :

Table I Intubation group I (29 patients)

Duration of intubation in days.			
<1	1-2	2-3	3-4
8 pts.	14 pts.	5 pts.	2 pts.

Table II Intubation group II (20 patients)

Duration of intubation in days.			
5-6	8-12	14-21	>21
3 pts.	11 pts.	3 pts.	3 pts.

II were treated in a respirator and 3 had cardiac arrest.

Within intubation group II there was clinically demonstrable damage to the larynx and subglottis in practically all patients, although no or negligible damage was found in those who had been intubated for 5 and 6 days. The severity of the laryngo-tracheal damage was to some extent dependent upon the duration of the intubation but this was not a rule without exceptions, as may be seen from Table III.

Among the cases designated ++ the vocal cords were found to be immobile in a position of adduction exactly like the appearance of "paralysis of the recurrent laryngeal nerve, but being in fact a sequel to fibrous changes in the articular and muscular apparatus of the vocal cords. Since no patient of group I had clinically demonstrable damage in the trachea and since the first 3 patients of group II did not have such damage or at least only very mild laryngo-tracheal changes, it may be concluded that naso-tracheal intubation may be maintained for 6-7 days without major risk of damage. If it is kept up longer it must be called prolonged intubation, which means a considerable risk of severe sequelae in the larynx and subglottis, possibly requiring treatment. In such cases, therefore it must be carefully contemplated whether instead of continuing the risk of prolonged

Table III Influence of the duration of intubation upon the severity of laryngo-tracheal sequelae

Sequelae			Sequelae		
No. of days	lar + subgl.	Trachea	No. of days	lar + subgl.	Trachea
5	0		11	+	
5	+		12	++	
6	+		12	++	
8	++	+	12	+	
8	++		14	++	
8	+++		15	0	
9	++		21	+++	
10	+++		26	+++	+
10	++	+	30	++	
10	++	+	36	+++	

+ Mild mucosal thickenings, granulation tissue.

++ Permanent, considerable stenosis of the lumen of firm, fibrous nature.

+++ Anatomical structures in the larynx are destroyed, larynx funnel-shaped, a few men passage to a greatly narrowed subglottis.

intubation it is not advisable to replace it by intubation through tracheostomy

REFERENCES

- Bergström, J. 1962. Laryngological aspects of the treatment of acute barbiturate poisoning. *Acta Otolaryng* (Stockh.), Suppl. 173.
- Dann, W. & Zwerghaus, E. 1952. Laryngeal complications after longvarig endotracheal intubation of narkotisk forgiftede patienter. *Nord Med* 48 1683.
- Fearon, B., MacDonald, R., Smith, C. & Mitchell, D. 1966. Airway problems in children following prolonged endotracheal intubation. *Ann Otol* 75 603.
- Hansen, M. & Jørgensen, S. 1968. Tracheostomy and prolonged nasotracheal intubation. *Danish Med Bull* 15 33.
- Halach, D. J. 1968. Prolonged nasotracheal intubation in infants and children. *Lancet* i 1273.
- Hedden, M., Ersoz, C. J., Donnelly W. & Safer P. 1969. Laryngotracheal damage after prolonged use of orotracheal tubes in adults. *JAMA* 207 703.
- Larsen, H. C. A. 1936. *Management of life-threatening pharyngitis*. London.
- Lewy R. B. & Sibbert, J. W. 1951. Tracheostomy in barbiturate poisoning. *Amer Pract* 2 257.
- Tyrlff, B. & Toa, M. 1969. Nasotracheal intubation in acute epiglottitis. *Acta Otolaryng* (Stockh) 68 363.

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TREATMENT OF ACUTE EPIGLOTTITIS IN CHILDREN BY LONG-TERM INTUBATION

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Abstract A description of the treatment of children with acute epiglottitis by long-term intubation is given. The results of treating 39 children by intubation are compared with the results of treating 51 children by tracheostomy.

Endotracheal intubation is now accepted as an alternative to tracheostomy in the treatment of acute epiglottitis. From November 1969 to November 1971 at the Central Hospital in Västerås, 39 children with acute epiglottitis were treated by endotracheal intubation. In this lecture we present our experiences with this method and compare it with 51 epiglottitis children treated by tracheostomy during the time 1962 to October 1969. During the whole of this period there was a total of 116 cases of which 51 were tracheostomized, 39 treated with intubation and the remaining 26 received only medical treatment.

There has been a successive increase in the number of epiglottitis cases per year (Fig. 1). Of the 90 cases who had been either tracheostomized or intubated 38 were girls and 52 boys. Age distribution is shown in Fig. 2. 79% of the patients are in the age group 2-6 years. All the patients, both tracheostomized and intubated, have received antibiotics and steroids, first by injection and afterwards orally. Large doses of Hydrocortisone were given up to approx. 1 gram on the first day. Afterwards Prednisolone in decreasing doses

was given. Average hospitalization for tracheostomized patients: 6.5 days, average time with tracheostomy 2.7 days. Average hospitalization for intubated patients 3.9 days, average time with tube, 1.5 days.

The longest time with a tracheostomy cannula was 6 days and the shortest, 1 day. The longest time with a tube, 2.5 days and the shortest, $\frac{1}{2}$ day.

Complications

We have had very few serious complications in association with tracheostomy. No deaths have occurred. All the tracheostomies were carried out under intubation anaesthesia. The complications we have had appear in Table 1 and from this it can be seen that in 17 of the 51 tracheostomized cases, 39% some form of complication occurred. Serious complications have only occurred, however in 2 cases, namely the bilateral pneumothorax and the granuloma in the trachea. In the first of these the intubation was extremely difficult and the anaesthetist never managed to get the tube past the vocal cords and the tracheostomy must be regarded as an emergency procedure. In the second case, with stridor approx. 1 month after tracheostomy direct laryngoscopy revealed a granuloma the size of a pea on the anterior wall of the trachea. Since this was removed the patient has been free of symptoms.

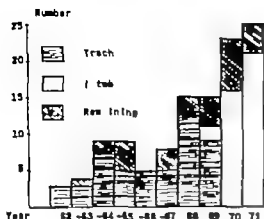


Fig 1 Number of children with acute epiglottitis per year from January 1962 to November 1971

In the 39 intubated cases no serious complications occurred as can be seen from Table II. There was, however, some form of minor complication in 15% of the cases. Among the 39 cases of intubation 1 death occurred which, however, had nothing to do with the method as the patient had already suffered a cardiac arrest before he was intubated.

Follow up

The intubated patients have been controlled after approx. 2 weeks by an indirect laryngoscopy and 34 of the 39 cases have been studied by direct laryngoscopy under anaesthesia between 1 and 11 months after the intubation treatment. In none of these cases was any remaining abnormality found in the epiglottis, larynx, or subglottically.

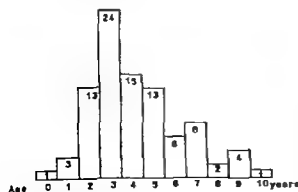


Fig 2 Age-frequency (90 patients).

Table I Complications with tracheostomy

1 Bilateral pneumothorax	1 case
2 Mediastinal emphysema	2 cases
3 Subcutaneous emphysema	3 cases
4 Small incision of posterior wall of trachea	1 case
5 Patient decannulated himself	1 case
6 Granuloma in trachea	1 case
7 Stridor and tendency to stenosis 1-2 months after tracheostomy	1 case
8 Disfiguring scars	7 cases

Total number of complications in 51 tracheostomized patients: 17

Details of technique

We feel that it is very important to lay advance plans for intubation treatment. It is obvious that good cooperation must exist between the departments of anaesthesia and E.N.T. During the time we intubate the child we have always an E.N.T. surgeon ready to do a tracheostomy if necessary. After the intubation an X-ray is taken immediately on the operation table. The child is anaesthetized with oxygen followed by N_2O/O_2 /fluothane and intubated without the help of muscle relaxants. Intubation is carried out first orally in order to judge the size of the tube and a tube is then selected for nasal intubation 0.5 or 1.0 mm smaller than that which easily passes through the larynx. It is important to stress that the tube should be as small as possible, without producing any respiratory obstruction. One of the big problems is sedation of the child. For this we use large doses of Diazepam which we give intravenously before the child is awake. We treat the child in an ultrasonically humidified tent and it is important to stress that the

Table II Complications with intubation

1 Coughed up tube	1 case
2 Extubated himself	1 case
3 Rapidly resolving atelectasis of the right upper lobe due to the tube being too far down	4 cases

Total number of complications in 39 intubated patients: 6

humidification must be perfect with these small tubes. The tubes are made from poly vinyl chloride, manufactured by Portex.

CONCLUSION

We believe that intubation treatment has obvious advantages over tracheostomy for the treatment of acute epiglottitis in children, for the following reasons:

1. It eliminates the risks for pneumothorax, mediastinal emphysema, subcutaneous emphysema etc. together with the other complications of tracheostomy.
2. It decreases risk for tracheal stenosis.
3. It avoids unsightly scars in the throat.
4. It reduces the period of hospitalisation, in this study on an average from 6.5 to 4 days.
5. Problems with decannulation, especially with small children, disappear.

Intubation has also some disadvantages. There is a risk of damage to the larynx and trachea. This would seem to be very small

in acute epiglottitis as the child is intubated for a short time only. We do not use a cuffed tube and no respirator is necessary. It is also important to stress that the tube should be as small as possible. It is somewhat more difficult to suck out the lungs through a small tube but with some experience this is by no means impossible. It is necessary to have good monitoring of the children and they are most suitably treated on an intensive ward with availability of an E.N.T. and an anaesthetic specialist.

Since this material was collected 10 further cases of acute epiglottitis have been treated in Västerås of which 7 required intubation. 4 of these have been directly laryngoscoped and found to be quite normal afterwards. The total of intubation treated patients is now 47 of which 38 have been checked afterwards.

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NASOTRACHEAL INTUBATION INSTEAD OF TRACHEOTOMY IN ACUTE EPIGLOTTITIS IN CHILDREN

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Abstract 30 children with typical, severe, acute epiglottitis were treated by intubation of 8-192 hours duration, mean 42 hours, instead of by tracheotomy. The method, results, and combinations of intubation are described. The results were good, complications few and negligible, and no damage to the larynx occurred. Since all children have to be intubated anyway in order to be tracheotomized, and since the critical period of airway obstruction lasts a maximum of 48 hours, the tube may just as well be left and tracheotomy avoided.

In acute epiglottitis immediate measures are often needed to secure the respiration. This was previously done by tracheotomy. In 1965 intubation was introduced in the Gentofte Hospital (Buchmann & Risker 1967) and in 1967 in the Glostrup Hospital (Triff & Tos, 1969). Since then tracheotomy has not been used in acute epiglottitis in these two hospitals. The material from Gentofte counts 10 cases, that from Glostrup 20 intubated children with typical, severe acute epiglottitis.

The youngest child was 1 year of age, the oldest 12. 17 were under 2 years, 7 between 3 and 4. The duration of symptoms before admission was more than 24 hours in only one case (Table I), that of respiratory embarrassment in most cases 2-6 hours. Many children were admitted in an extremely poor condition 7 with distinct cyanosis. 17 had to be intubated immediately (Table I).

Twenty-five children had nasotracheal intubation, 5 orotracheal under general anaes-

thesia using smooth polyvinyl chloride tubes. The intubation was done in the operating theatre of the ENT Dept. with readiness for tracheotomy and bronchoscopy. As a rule it could be carried out at first attempt. By Melstosch laryngoscope and the tube the epiglottis was pressed forward. The posterior edge of the arytenoid region was then visible, and the tube could be passed. The tubes were as thin as possible so as not to irritate the vocal cords and from the outset allow respiration outside the tube. The patients were placed in an Air Shield croupette. The object was to extubate as soon as possible, even though oedema of the epiglottis remained. The critical period of acute epiglottitis is the first 24 hours thereafter the oedema starts subsiding. The intubation lasted in most cases for 25-36, mean 42, hours (Table I). Two patients could be extubated after 8 hours, one was intubated for 192 hours. This latter patient had acute epiglottitis, but also subglottic laryngitis. After 72 hours' intubation extubation was tried, but the patient had to be re-intubated for 48 and thereafter again for 72 hours.

Complications and symptoms following extubation were few and negligible. Constant stridor was observed in 7 cases, lasting for 1-2 hours in three and for 3-6 hours in three. Only one patient had mild, constant stridor for 24 hours. This patient had been intubated for 48 hours. In 5 patients there was inter-

Table I Duration of symptoms and of respiratory embarrassment before admission. Time of intubation after admission and duration of intubation in hours

	Number of patients in each time interval											Mean time in hours
	Immedi- ately	Time intervals in hours										
		1	2-3	4-6	7-12	13-24	25-36	37-48	60-67	III	19 ^a	
Duration of symptoms				3	11	15		1				16
Duration of respiratory embarrassment			8	17	2	3						6
Time of intubation	17	5	5	1	2	4	10	8	4	1	1	1
Duration of intubation					2	4	10	8	4	1	1	42

mittent stridor mainly during sleep, as a rule for 24 hours, but in one case for 4 days. Stridor following extubation was due to the still swollen epiglottis, hardly to oedema of the vocal cords or of the subglottis, caused by the intubation. The mean intubation period in patients with stridor had been 41 hours, in those without 43 hours.

Hoarseness was found after extubation in 9 cases, in 2 of them for 3 and 8 days respectively. Both had been intubated for only 24 hours. In the remaining 7 cases the hoarseness lasted 1-48 hours. Hoarseness following extubation must be due to oedema of the vocal cords caused by the tube. The mean intubation period in patients with hoarseness was 59 hours, without hoarseness 35 hours. The Glostrup series was examined 1-4 weeks and the Gentofte series 1-12 months after discharge. In all cases the vocal cords and voice were normal.

During energetic antibiotic therapy the Gentofte series, but not the Glostrup series, was treated with Actocortin. The mean intubation period in 10 Actocortin-treated children was 33 hours, in 20 non-Actocortin-treated ones 39 hours. The time it took the epiglottis to become normal was 6 days in both series. Actocortin is hardly necessary and presumably has no convincing effect upon the normalization of the epiglottis. At least intubation should not be delayed while waiting for the questionable and slow effect of Actocortin, as the respiratory embarrassment may very rapidly get worse.

Prolonged intubation is, and should still be feared by otologists because of its damage to the larynx. Severe laryngeal damage is often reported following prolonged intubation. However it is difficult to define what prolonged intubation is, as this does not depend only upon the intubation period, but also on many other factors. Nasotracheal intubation as administered in the present material, can hardly be considered prolonged intubation, and possesses several advantages above tracheotomy: (1) Children are in a very poor condition, and the airway has to be secured immediately. This can only be done sufficiently well by intubation. (2) Even though the child has been intubated, it is still very ill, so that acute tracheotomy with its well-known complications during and after the operation, especially common in children, is not indicated. (3) Extubation problems are less serious than decannulation problems. (4) The psychic trauma is not greater in intubated than in tracheotomized children.

REFERENCES

- Bachmann, H. & Rikner, N. 1967. Laryngitis stridula hos børn behandlet med langvarig intubation. *Nord Med* 77: 642.
 Triff, B. & Tox, M. 1969. Nasotracheal intubation in acute epiglottitis. *Acta Otolaryng* (Stockh.) 66: 368.

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TREATMENT OF STENOSIS OF THE TRACHEA

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Abstract A closed tubulation-technique is described for the treatment of patients with stenosis of the trachea. The results of treatment in 29 patients with stenosis of the trachea—induced mainly by prolonged intubation—are reported.

The operation is performed through an external incision. A tube of silicone is enclosed in the trachea and sutured to the tracheal wall. In this way normal respiration through the upper respiratory tract is possible. In 22 patients this treatment was a success. In 7 patients the tube had to be removed because of respiratory failure and stagnation of secretion.

The method seems to be suitable in all patients with stenosis and malacia of the trachea except in patients with respiratory failure and stagnation of secretion. A transverse resection may be preferable in stenosis of limited size in the upper trachea.

This paper will be published in its entirety elsewhere.

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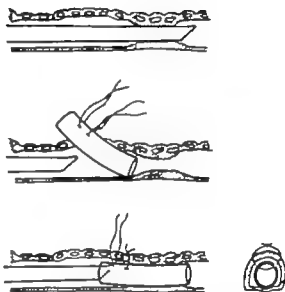


Fig 1 Schematic drawing of the procedure. (Upper) ventilating bronchoscope introduced through the stenosis of the trachea. (Centre) insertion of the tube. (Lower) tube in position. (Right) transversal section of the trachea with tube in position, showing suture.

ANAESTHESIA IN OPERATIONS FOR LARYNGEAL AND TRACHEAL STENOSIS

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Abstract. The authors describe anaesthesia methods in 61 operations for the correction of laryngeal and/or tracheal stenosis. Methoxyflurane (Penthrane®) with large doses of clonitiniol chloride (Sömac®) and diazepam was found to be the most practical and was the anaesthesia mainly used.

Since 1966, 61 operations for the correction of laryngeal and/or tracheal stenosis have been performed in the Helsinki University Otolaryngological Hospital. The technique of these operations was previously described in detail by Grahne (1971) and by Grahne & Poppius (1972). The permanent chronic stenoses can be found on three different levels: in the larynx, in the cervical and in the thoracic trachea.

Prostheses according to Aboulker (1962, 1968) and Aboulker et al. (1966) and Aubry & Bourdon (1960) were used. At the very beginning of the series a prosthesis according to Clerici also was tried. At first we had difficulties in finding a convenient method for anaesthesia.

Choice of anaesthesia

There were several problems. It was important not to reduce the already narrow airway with an endotracheal tube. It was also necessary to maintain the spontaneous respiration of the patient during various phases of the operation. In the laryngeal stenosis cases we could by pass the constricted area by performing a

tracheotomy and inserting an endotracheal tube below the stenosis.

In the operations of the cervical and intrathoracic trachea we encountered much greater problems. In these cases there was no room for an endotracheal tube. We had to find some other practicable method of anaesthesia to supplement the effect of the premedication and *local infiltration anaesthesia*.

The anaesthesia methods in the various operations are shown in Table I. At first we used a combined nitrous-oxide halothane anaesthesia. This combination however did not permit the use of vasoconstricting agents in connection with the necessary local anaesthesia. The patient had to breath spontaneously on several occasions because there was no room for the endotracheal tube or for artificial ventilation.

Next choice was methoxyflurane (Penthrane®) a long-lasting anaesthetic which preserves the spontaneous respiration even in deep levels of surgical anaesthesia. Penthrane also has fewer side effects than halothane when administered simultaneously with vasoconstrictors.

Many problems were caused by the great sensitivity of the endolaryngeal and particularly of the subglottic region with the subsequent strong cough reflex. This cough reflex could not be controlled with halothane or methoxyflurane. Muscle relaxants could not

Table I Anaesthesia in operations for tracheal and/or laryngeal stenosis

Operation	Anaesthetic		Other I.v.	Hal.	Mor	Σ
	Local	SO- Dzp				
Clerici	3	—	1	11	—	15
Aubry	1	2	2	2	3	12
Aboulker	3	5	6	2	7	29
Intrathor	1	2	—	—	—	5
Σ	8	9	8	15	10	41

be used to any great extent because of the occasional need for spontaneous respiration. The primary operation was always begun with a tracheotomy which already indicated sedation.

We then administered diazepam intravenously with a total dosage of 10–70 mg, which already gave a very satisfactory sedation. However diazepam did not help beyond the tracheotomy mostly performed under local anaesthesia, because the posterior wall of the larynx needs a much stronger anaesthesia. Nor was diazepam capable of eliminating cough reflexes during the operation in the endolarynx.

During the postoperative phase of previous laryngeal and tracheal operations we had observed the very good effect of clobutinol chloride (Silomat®) in controlling the cough reflex. The dose of clobutinol chloride, however was much larger than that needed in most diseases with cough symptoms. Our doses varied between 20–80 mg given in intermittent I.v. injections of 5–10 mg. We found that clobutinol chloride, even when given simultaneously with diazepam reduced the cough reflex very effectively without depress-

ing the respiration. In those cases where the respiration was secured by an endolaryngeal tube we were able to supplement the anaesthesia by using methoxyflurane or a muscle relaxant. These drugs could not be used, when it was necessary to have good spontaneous breathing during the operation within the trachea and during the positioning of the prosthesis.

After the Aboulker prosthesis had been placed in position, the height of its upper end was checked by direct laryngoscopy carried out by the anaesthetist. When the Aboulker prosthesis was correctly placed, anaesthesia was continued through the silver cannula until the end of the operation. Diazepam-Silomat combination was also very effective for the final control laryngoscopy.

REFERENCES

- Aboulker P 1962. Chirurgie de la trachée. *Probl Actuels Otolaryng* p 273
 — 1968. Traitement des sténoses trachéales. *Probl Actuels Otolaryng* p 275
 Aboulker P., Sterkers, J. M., Demaldent, J.-E. & Sarron, P 1966. Modifications apportées à l'intervention de Rethl. Intérêt dans les sténoses laryngo-trachéales. *Ann Otol (Par)* 83 98.
 Aubry J & Bourdon, J 1960. Contribution à l'étude des sténoses laryngées cicatricielles. Thesis. Paris
 Grahne, B. 1971. Operative treatment of severe chronic traumatic laryngeal stenosis in infants up to three years old. *Acta Otolaryng (Stockh)* 71 134
 Grahne, B. & Popplus, H. 1973. Transtracheal cervical approach for operating on severe intrathoracic tracheal stenosis following assisted respiration. *Acta Otolaryng (Stockh)* 75 64

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EXPERIMENTAL TRACHEAL WALL INJURY

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Abstract. A plan for experimental analysis of tracheal wall injuries in rabbits is presented. Preliminary results after different types of injuries to the tracheal mucosa are given.

During recent decades the problem of tracheal wall injury has been discussed in several clinical and experimental investigations (Ekedahl & Laage Hellman, 1967 Harrison & Tonkin, 1966 1968 Lindholm, 1969 Miller & Sethi, 1970 Hilding, 1971). None of these papers have, however solved the fundamental problem of the effects of different parameters such as wall pressure, intubation time, size of tube, etc., on the tracheal wall. It must be assumed that these factors, either in isolation or together may cause irreversible changes in various wall structures.

Against this background we have tried to analyse the degree of tissue injury from different points of view. Fig. 1 is a schematic representation of the methods used in this study.

Lesions of the tracheal wall in rabbits have been induced experimentally under defined conditions and analysed morphologically according to a method described by Björkerud (1969 1972). The types of injury induced were superficial with either small or large surface area. The animals were sacrificed after intervals varying between 24 hours and 4 weeks. Numerous leucocytes and goblet cells were present in and near the damaged zone 1 week after the trauma. Ciliated epithelial cells were found in uninjured parts of the trachea but absent in

the injured region. Three weeks after the trauma tortuous capillaries with varying diameters, monocytes, and focal necrotic areas were found. Characteristic features of the necrotic areas were a central elevated region with necrotic cells and leucocytes. Both the necrotic and the immediately surrounding zone were devoid of epithelium. Thus, a very small trauma to the tracheal mucosa is followed by such protracted changes as subchronic inflammation, focal areas with necrosis, and incomplete regeneration of the epithelium. Further studies are needed to elucidate the repair capacity of the tracheal wall in more detail and which factors may promote or prevent the repair of the tissue.

The tracheal vascular bed has been visualized by means of intra-arterial perfusion of Indian ink in non-intubated and intubated rabbits. The intubation time has been 90 minutes. The wall pressure used in this study was 20 mmHg (to be published). The normal architecture of the vascular bed in the rabbit is dominated by the venous plexa situated in the submucosa between the cartilage rings. These plexa are connected by arcades of vessels crossing the rings. There is also a very fine net of blood vessels in the mucosa. In microangiograms from rabbits sacrificed with the cuffed tube still in the trachea, most of the vessels are blocked and only a few arterioles are found in the submucosa. In the animals sacrificed 40 minutes after extubation an almost normal architecture of the

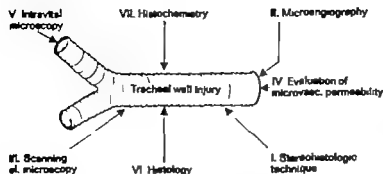


Fig 1 Schematic representation of the methods.

arterioles and venules was found. The microvessels in the mucosa were poorly filled, however. These findings indicate vascular injury to the capillary network.

Only preliminary results with a few methods are here discussed. Other ways of attacking these problems (as was indicated in the figure) are presently being studied. We hope that further studies along these lines will clarify the mode of reaction of this tissue.

REFERENCES

- Björkerud, S. 1969 Reaction of the aortic wall of the rabbit after superficial longitudinal mechanical trauma. *Virchow Arch Path Anat* 347: 197.
- 1972 Preparative, staining and microscopic techniques for the study of whole artery segments. *Atheroscl* 15: 147.
- Ekdahl, C. & Lange Hellman, J.-E. 1967 Tracheal changes after tracheostomy. *Acta Otolaryng* (Stockh.), Suppl. 224: 40.
- Hanson, H., Hansson, P. & Nilsson, K. 1972 Cuff pressure at endotracheal intubation. *Acta Otolaryng* (Stockh.) 75: 391.
- Harrison, G. A. & Tonkin, J. F. 1968, Prolonged (therapeutic) endotracheal intubation. *Brit J Anaesth* 40: 241.
- Hilding, A. C. 1971 Laryngotracheal damage during intratracheal anesthesia. *Ann Otol* 80: 563.
- Lindholm, C.-E. 1969 Prolonged endotracheal intubation. *Acta Anaesth Scand*, Suppl. 33.
- Miller, D. R. & Seth, G. 1970 Tracheal stenosis following prolonged cuffed intubation. Causes and prevention. *Ann Surgery* 171: 283.
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EXPERIENCE WITH A NEW OROTRACHEAL TUBE

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Abstract. A preshaped tube which conforms to the contour of the airway exerts less pressure against the posterior wall of the larynx than a conventional tube. Clinical evidence is presented which shows that lesions in the posterior part of the larynx are significantly less pronounced following prolonged oro-tracheal intubation with preshaped tubes than with conventional tubes.

Without exception, observations have shown that conventional straight or slightly curved oro- or nasotracheal tubes exert a considerable pressure against the posterior wall of the larynx and the anterior wall of the trachea. The shape of a conventional tube is far from ideal and when it is reshaped according to the anatomical structures, when introduced into a patient, it gives rise to undue pressure necrosis and ulcerations. It is to be expected that these kinds of lesions can be avoided or greatly reduced if a preshaped tube is used. A tube which conforms to the airway is now available and is manufactured by Portex Ltd. (Fig. 1).

A comparison has been made between 69 adult patients (Series I) treated with indwelling conventional oro-tracheal tubes for >24 hours (mean 32 hrs) and 45 adult patients (Series II) treated with indwelling preshaped oro-tracheal tubes for >24 hours (mean 44 hrs). Both groups were otherwise treated in the same manner in the same intensive care units, and by the same staff. All patients were carefully examined by repeated indirect or direct laryngoscopies following the period of intubation. Acute damage in the larynx was classified according to previously described principles (*Acta*

Anaesth Scand Suppl. 33 p 53) The results are given in Table I.

The lesions healed within 15 days in 23% of the patients in Series I and in 60% in Series II within 30 days in 88% in Series I and in 91% in Series II. Delayed healing with associated granuloma formation occurred in 33% in Series I and in 9% in Series II and healing times longer than 30 days occurred in all of these patients.

No voice changes or voice changes lasting less than 1 week were noted by 66% of the patients in Series I and by 74% in Series II changes lasting from 1 to 3 weeks were noted by 16% of the patients in Series I and by 17% in Series II. Voice changes lasting 1 month or more were reported by 18% of the patients in Series I and by 9% in Series II.

Airway obstruction in the larynx following extubation was observed in 1 patient in Series I. No such difficulties were experienced by patients in Series II.

The carina to tube distance was measured in routine chest radiographs. In women this distance varied between 2.7 and 5 cm (mean 3.3 cm) in men between 2 and 5.4 cm (mean 4.1 cm). These figures are valid for the recommended tube sizes, e.g. in women 7.5 mm ID in men 8.0 or 8.5 mm ID.

This investigation confirms that lesions in the posterior part of the larynx are significantly less pronounced when preshaped oro-tracheal tubes are used instead of conventionally shaped tubes.

In my opinion it is desirable to use a pre-

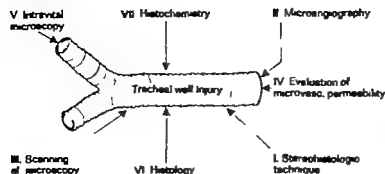


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REFERENCES

- Björkerud, S. 1969 Reaction of the aortic wall of the rabbit after superficial longitudinal mechanical trauma. *Virchow Arch Pathol Anat* 347 197
- 1972. Preparative, staining and microscopic techniques for the study of whole artery segments. *Atheroscl* 15 147
- Ekelöf, C. & Laage-Hellman, J. E. 1967 Tracheal changes after tracheostomy. *Acta Otolaryng* (Stockh.), Suppl. 224 40.
- Hanson, B., Hanson, P. & Nilsson, K. 1972. Cuff pressure at endotracheal intubation. *Acta Otolaryng* (Stockh.) 75 391
- Harrison, G. A. & Tonkin, J. P. 1968 Prolonged (therapeutic) endotracheal intubation. *Brit J Anaesth* 40 241
- Hilding, A. C. 1971 Laryngotracheal damage during intratracheal anaesthesia. *Ann Otol* 80 565
- Lindholm, C. E. 1969 Prolonged endotracheal intubation. *Acta Anaesth Scand* Suppl. 33
- Müller, D. R. & Seth, G. 1970. Tracheal stenosis following prolonged cuffed intubation: Cause and prevention. *Ann Surgery* 171 283
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CUFF PRESSURE AT ENDOTRACHEAL INTUBATION

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Abstract. A preliminary investigation on the tracheal wall pressure from endotracheal tube cuffs is reported. A simple technique for calculation of tracheal wall pressure is presented. A limited clinical material has been analysed regarding cuff inflation volume, cuff pressure and resulting wall pressure.

It is well known that cuffed endotracheal tubes can produce varying degrees of local tissue damage (Adrian & Phillips, 1957; Cooper & Grillo 1969; Hilding, 1971).

As a background to further experimental studies on tissue injury produced by such cuffs the pressure load on the tracheal wall has been analysed. Various methods have been devised giving a direct or indirect measurement of the actual wall pressure (Adrian & Phillips, 1957; Carroll et al. 1969; Hilding, 1971; Knowlson & Basset, 1970). All of these methods include some kind of arrangement either on the tube or in the tracheal wall.

One of the main aims of the present study was to evaluate the pressure load at routine use of cuffed rubber tubes (Rüsch). A method was therefore required that could be used without previous information to the staff working daily with induction of anaesthesia and intubation.

By measuring the actual cuff inflation volume and pressure with the tube in the trachea and the corresponding volume and pressure situation with the free tube it is pos-

sible to obtain a fairly accurate value of the lateral pressure caused by the cuff. This hypothesis was tested and also confirmed in model experiments on autopsy material.

Fig. 1 shows the increasing cuff pressure with successive inflation of the intratracheal cuff and of the free cuff. The calculated resulting wall pressure is given on the abscissa. The steep rise in the later part of the wall pressure curve is important to emphasize.

The same method can be used to elucidate minimum occlusive volume (MOV) and corresponding minimum occlusive wall pressure (MOWP) for different intratracheal pressures. Theoretically minimum occlusive wall pressure should equal or just exceed the intratracheal pressure (Adrian & Phillips, 1957; Knowlson & Basset, 1970).

Fig. 2 shows the result of measurements on minimum occlusive wall pressure for two tube dimensions in the same cadaver trachea. With this set-up the necessary wall pressure exceeds the intratracheal pressure with about 10 mmHg. A pronounced difference in total cuff pressure is not reflected in the calculated wall pressure. The minimum occlusive wall pressure is compared with an expected arteriolar pressure of 40 mmHg.

With this "calculated wall pressure technique" a limited clinical material has been analysed. Fig. 3 shows the registered total cuff pressures and the resulting calculated wall pressures. It is obvious that routine intubation

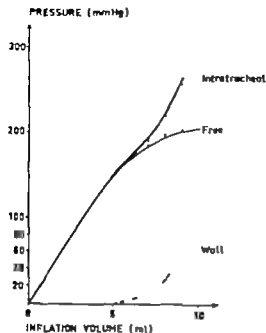


Fig 1 Cuff pressure-volume relation for the free and the intratracheal cuff and the resulting wall pressure

results in a high intracuff pressure with a great variability. It should be emphasized, however, that there is no direct correlation between this total cuff pressure and the resulting wall pressure nor is there a direct correlation between the cuff inflation volume and the wall pressure.

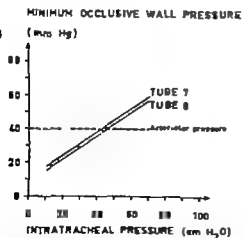


Fig 2 Relation between the minimum occlusive wall pressure and the tracheal pressure for two tube dimensions in an experimental model.

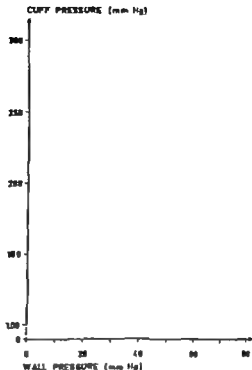


Fig 3 Total cuff pressure and calculated wall pressure in a clinical material.

REFERENCES

- Adriani, J & Phillips, M. 1957 Use of the endotracheal cuff: Some data pro and con. *Anesthesiology* 18 1.
- Carroll, R., Hedden, M & Safar, P. 1969 Intratracheal cuffs: Performance characteristics. *Anesthesiology* 31 275.
- Cooper, J. D. & Grillo, H. C. 1969 The evolution of tracheal injury due to ventilatory assistance through cuffed tubes. A pathologic study. *Ann Surg* 169 334.
- Hilding, A. C. 1971 Laryngotracheal damage during intratracheal anesthesia. *Ann Otol* 80 565.
- Knowlton, C. T. G. & Basset, H. F. M. 1970. The pressures exerted on the trachea by endotracheal inflatable cuffs. *Brit J Anaesth* 42 834.
- Lindholm, C. E. 1969 Prolonged endotracheal intubation. *Acta Anesth Scand Suppl.* 33.

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INTERACTION BETWEEN THE UTRICLES AND THE HORIZONTAL SEMICIRCULAR CANALS

II. Unilateral Selective Section of the Horizontal Ampullar and the Utricular Nerve followed by Tilting around the Longitudinal Axis

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Abstract. Selective unilateral section of the horizontal ampullar nerve and the utricular nerve was performed. By tilting the animals around their longitudinal axis, the alterations in the horizontal nystagmus were studied. It was shown that these alterations were released from the intact utricle. An increase in utricular activity due to tilting toward the operated ear resulted in a positive triggering of the nystagmus from the ipsilateral horizontal semicircular canal. A decrease in its activity by tilting toward the nonoperated ear inhibited the nystagmus. The cause of these findings is discussed.

In a previous article (1972) the authors have shown that tilting of an animal around its longitudinal axis, after selective unilateral section of the horizontal ampullar nerve, results in a characteristic alteration in the frequency of nystagmus. Tilting toward the operated side increased the frequency and tilting toward the nonoperated side caused inhibition of nystagmus. This has resulted in two questions which will be studied in this paper:

1. Does the alteration in nystagmus depend on variation of activity in the utricle?
2. If so what effect has the utricle on the ipsilateral horizontal semicircular canal?

MATERIAL AND METHOD

Sixty-two adult cats were used for the experiments. The operation, stimulation, and recording techniques are the same as those described in a previous article

RESULT

All the cats were first subjected to selective unilateral section of the horizontal ampullar nerve, and manifested nystagmus toward the contralateral side. This nystagmus did not change perceptibly when the utricular nerve was also cut. If the animals were tilted toward the operated side 41 of them showed an increase in nystagmus frequency often from about 17 up to 31 beats/10 seconds. If the nystagmus had previously been somewhat irregular in shape, it now became much more regular as regards both frequency and amplitude. If instead the cats were tilted toward the nonoperated side we found in all of them alterations in nystagmus. In 48 cases there was a total inhibition of nystagmus, which occurred either immediately (12 cases) or slowly after 10-20 seconds (7 cases) with a continuous fall in frequency. Initially 29 cats showed an increase in the frequency of nystagmus, with 5-14 beats/10 seconds, later followed by total inhibition (Fig. 1). Fourteen animals had also an initial increase in frequency from about 17 up to 41 beats/10 seconds, while the amplitude decreased continuously from 2-3 cm down to 1-2 mm, however without total inhibition of nystagmus. Thirty-six cats showed an "ideal" reaction when tilted in either direction, i.e. facilitation during tilting toward the operated ear and inhibition toward the non-operated side.

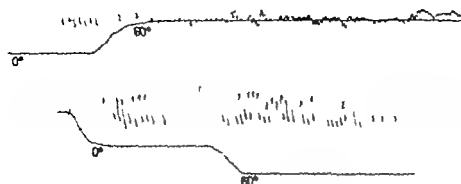


Fig. 1. Consecutive curves from a cat after selective unilateral section of the left horizontal ampullar nerve and the left utricular nerve. The upper curve shows

the electronystagmography and the lower curve the tilting of the table. Tilting to the right upwards, to the left downwards.

DISCUSSION

We have created a system with only the horizontal semicircular canal and the utricle on one side intact (Fig. 2). It has been clearly demonstrated that variations in the discharge frequency of the intact utricle caused by tilting influence the ipsilateral horizontal semicircular canal or its reflex arc. An increase in utricular activity which occurs during tilting toward the operated ear is consequently triggering the reflex positively for ipsilateral nystagmus (Fig. 3). This also explains the findings (Fluor & Mellström, 1970) that electrical stimulation of the midlateral lar area can result in ipsilateral horizontal nystagmus, without this being a result of the current spreading through the endolymph to the horizontal ampulla or its nerve. Consequently every depolarization of this utricular area, whatever its cause, facilitates the activity of the ipsilateral horizontal canal or its reflex arc.

On the other hand, a decrease in the activity in the intact utricle which happens during tilting toward the nonoperated side causes a definite inhibition of nystagmus (Fig. 4). In analogy with the argumentation referred to above is this finding due to the fact that a decreased utricular activity lowers the triggering of the horizontal nystagmus down to

complete inhibition. These results correspond to similar findings in labyrinthectomized rabbits by Jongkees & Philipszoon (1964).

This inhibition can evidently develop in different ways, either directly or via an initial increase in frequency. How this is brought about is not known. It may be through direct anatomical anastomosis between the utricle and the horizontal semicircular canal found by Owada et al. (1960) but there are also signs indicating that it takes place all the way up in the oculomotor system being a result of the integrating activity in this system described by Yamanaka & Bach-y-Rita (1968, 1970). The activity of the reticular formation and the wakefulness of the cats are certainly also of great importance.

ZUSAMMENFASSUNG

Selective unilaterale Absehnungen der N. ampullaris canalis horizontalis und N. utricularis wurden vorgenommen. Durch Kippung der Tiere um ihre longitudinale Achse wurden die Veränderungen des horizontalen Nystagmus studiert. Es wurde gezeigt, dass diese Veränderungen vom intakten Utriculus ausgelöst werden. Eine Erhöhung seiner Aktivität durch die Kippung in Richtung des operierten Ohres ergab eine Steigerung der Nystagmusgeschwindigkeit, ausgelöst vom ipsilateralen horizontalen Bogengang. Eine Herabsetzung der Aktivität durch Kippung in Richtung des nicht operierten Ohres ergab eine Inhibition des Nystagmus. Der Grund dafür wurde diskutiert.

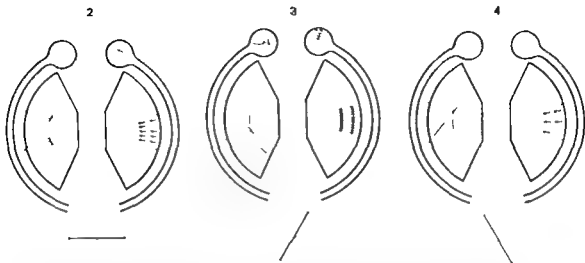


Fig 2 Schematic picture of the utricles and horizontal semicircular canals in horizontal position after section of the left horizontal ampullar nerve and left utricular nerve. The arrows indicate the orientation of the hair cells, i.e., the direction in which they increase their discharge frequency. The number of arrows symbolizes discharge frequency. The line below the picture symbolizes the direction of tilting.

Fig 3 Schematic picture of the utricles and horizontal semicircular canals after unilateral selective section of the left horizontal ampullar nerve and the left utricular nerve, and when the animal is tilted to

the left. Arrow indication same as in Fig. 2. The number of arrows symbolizes discharge frequency. The line below the picture symbolizes the direction of tilting.

Fig 4 Schematic picture of the utricles and horizontal semicircular canals after unilateral selective section of the left horizontal ampullar nerve and the left utricular nerve, and when the animal is tilted to the right. Arrow indication same as in Fig. 2. The number of arrows symbolizes discharge frequency. The line below the picture symbolizes the direction of tilting.

REFERENCES

- Flaar E. & Mellström, A. 1970. Utricular stimulation and oculomotor reactions. *Laryngoscope* 80 1701.
- Flaar E. & Slegbo, J. 1972. Interaction between the utricles and the horizontal semicircular canals. I. Unilateral selective section of the horizontal ampullar nerve followed by tilting around the longitudinal axis. *Acta Otolaryng (Stockh)* 75 17.
- Jongkees, L. B. W. & Philippon, A. J. 1964. Electro-nystagmography. *Acta Otolaryng (Stockh)*, Suppl. 189.
- Owada, K., Shizu, S. & Kimura, K. 1960. The influence of the utricle on nystagmus. *Acta Otolaryng (Stockh)* 52 215.
- Yamada, Y. & Bach-y-Rita, P. 1968. Conduction velocities in the abducens nerve correlated with vestibular nystagmus in cats. *Exp Neurol* 20 143.
- 1970. Relations between extraocular muscle contraction and extension times in each phase of nystagmus. *Exp Neurol* 27 57.

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THE ROLE OF THE PERILYMPH IN SEMICIRCULAR CANAL STIMULATION

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(Received June 21 1972)

Abstract The assumption of a mechanical function of the perilymph in vestibular reactions has been reiterated in the literature (Anliker M & v Buskirk, W 1971). The role of the perilymph in response of the semicircular canals to angular acceleration. *Acta Otolaryng* (Stockh.) 72 93). This assumption is essentially based on three premises: that the perilymph should meet a lower resistance than the endolymph because of the larger perilymphatic space, that the ampulla blocks perilymph movement by presenting one wall perpendicularly placed at the end of the perilymphatic canal and that this ampullary wall and the perilymphatic meshwork of loose connective tissue offer no resistance to perilymph movement. This paper presents criticisms of each of these premises and maintains that the assumption of a mechanical function for the perilymph cannot be supported or sustained by reference to them.

The assumption that the perilymph may play a role in the physiological stimulation of the sensory epithelium of the cristae ampullares is not new and has appeared occasionally in several publications and discussions. It might be argued that a structure like the loose connective tissue of the perilymphatic space, which is found in all higher animal species, might therefore be assumed to have a specific and perhaps an important function. However the discovery of the significance of the diffusion of water and electrolyte ions through the membranous walls of the inner ear for the equalization of osmotic pressure in endolymph

and perilymph might logically seem to be a satisfactory explanation for the presence of perilymph. The chemical constitution of this fluid is a result of ultrafiltration of water and ions from the blood vessels and corresponds to that of other extracellular fluids.

This low viscosity fluid is *not* however a freely movable fluid within the bony walls of the vestibular labyrinth. It is well known since the investigations by de Burlet (1921) that a relatively dense vascularized network of connective tissue turns the vestibular perilymphatic space into a spongy meshwork of varying density which must present an obstacle to the free flow of perilymph. In the cochlear perilymphatic scalae and in the vestibular cisterns surrounding the sacculus there are no such obstacles to an unimpeded perilymph movement. This arrangement is necessary in these regions for the transmission of soundwaves (de Burlet, 1921). This absence or presence of the meshwork of loose perilymphatic connective tissue is the important physical difference between the nature of the perilymphatic space in the vestibular part and that of the cochlear saccular part. These regions of the inner ear are separated by an incomplete dividing membrane, *Membrana limitans* (de Burlet, 1921).

The assumption of a mechanical function of the perilymph in vestibular reactions was first proposed by Reijto (1939) and later reiterated by Anliker & v Buskirk (1971). These

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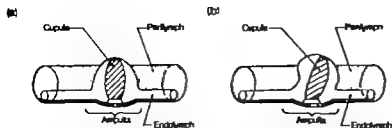


Fig. 1 (a, b) Conjecture of Anliker & v Buskirk (1971) as to possible mechanics of cupular deflection. The ampulla is considered to block perilymph movement by presenting one wall perpendicularly

placed at the end of the perilymphatic canal, as in (a). (b) The resulting cupular movement due to the presumed perilymphatic flow

latter authors essentially based their work on the following premises:

(1) Due to the larger size of the perilymphatic space, the perilymph should meet a much lower resistance from the effect of viscosity than the endolymph would experience in the narrow membranous canals.

(2) It was further assumed that the ampulla blocks the perilymph movement by presenting one wall perpendicularly placed at the end of the perilymphatic canal. The cross-sectional area of this wall is assumed to be approximately equal to the cross-sectional area of the ampulla (Fig. 1).

(3) It was also conjectured that the perilymphatic meshwork as well as the ampullary wall should offer no resistance to perilymph movement. The indentation of the wall which should then result from perilymph movement was assumed to cause the cupula to bend and to stimulate the hair cells.

Comments on the First Premise of Anliker and v Buskirk

To support their statement regarding the difference in cross-sectional area between the endolymphatic canal and the surrounding perilymphatic space these authors reproduce a picture from *Gray's Anatomy*. As scientific support for a theory which greatly deviates from generally accepted viewpoints, its acceptance is questionable.

It must be assumed that these authors would

not except their calculations to be valid only for the human vestibular apparatus. The conditions of the perilymphatic space is especially difficult to assess in this species due to the impossibility of acquiring temporal bone fixation without the postmortal changes that easily damage this tissue and also due to the frequent distortions by gas bubbles which develop during decalcification of the thick and dense human labyrinthine bone. It is presumed therefore that the results of Anliker and v Buskirk are meant to be applicable also to other animal species which have a perilymphatic and endolymphatic system like that of man. The perilymphatic tissue appears to be very thin in humans for the reasons mentioned, and even in other species the real density of this tissue might not be adequately realized without special methods for its demonstration.

Further it appears that these authors have not made any corrections of their value of the cross-sectional area of the perilymphatic space due to its specific configuration. This area is not a circular or ellipsoidal area as they have depicted their concept. The discrepancy between this hypothetical assumption and the cross-sectional area as it appears in reality is illustrated in Fig. 3a drawn from the measurements obtained from the pigeon's horizontal canal in Fig. 2. This drawing represents a cross-section through the endolymphatic and perilymphatic spaces for the purposes of estimating the friction these spaces provide to the fluid contained therein. To

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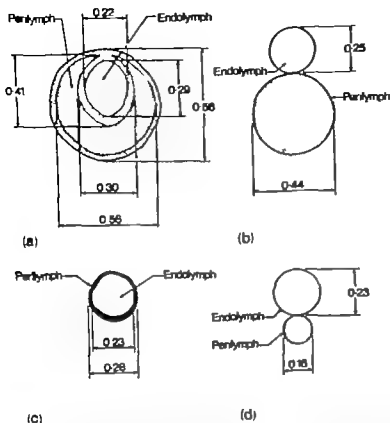


Fig. 3 Drawings of cross-sections through the horizontal canals of various animals. (a) A drawing of the pigeon's canal taken from Fig. 2; it shows the relative dimensions of the endolymphatic and perilymphatic lumens. (b) The conjecture of Anliker & v Buskirk (1971) as to the orientation of these two spaces in the pigeon. (c) Represents the actual situation

in a cat and it is seen that the perilymph is a thin ring around the endolymph. (d) Represents the associated model of Anliker & v Buskirk (1971) relative to the situation for the cat. It is clear from (b) and (d) that the Anliker & v Buskirk model of the endolymph-perilymph relation is not representative of the actual situation in the pigeon and the cat.

relatively large but in the cat the perilymph constitutes only a thin rim around the membranous canal (Fig. 3c and d). This information is easily obtained from the most precise investigation on this subject performed by Fernández & Valentinuzzi (1968). According to these measurements the diameter of the osseous horizontal canal is 0.28 mm and in this space the membranous canal has a diameter of 0.23 mm leaving a perilymphatic space of at most 0.025 mm in width surrounding the membranous canal. The perilymphatic space was even smaller in the vertical canals. If the perilymph is of the great importance ascribed to it by Anliker & v Buskirk, the cat

should have a correspondingly lower sensitivity to acceleration than either man or the pigeon. However the cat is one of the more commonly used animals for precise vestibular experiments and there has been no report that its sensitivity to acceleration is lower than in those species which have a much wider perilymphatic space. Further considering the relationship between changes in the cross-section and the radius of curvature of the membranous canals versus the body mass in different animal species as studied by Melvil Jones & Spells (1963), only the endolymphatic space seems to be relevant in this context in spite of the variations in perilymphatic space.



Fig. 4 Dark-field photograph and drawing depicting the angle between the horizontal canal and ampulla in the pigeon. The bony wall of the right and left horizontal ampulla and canal is peeled off from the

anterior-inferior side to show the insignificant part of the ampullary wall facing the canalicular perilymphatic space.

Comments on the Second Premise of Anliker and v Buskirk

In the schematic drawings by Anliker & v Buskirk (1971) (their figures 1 and 4) the important point which is emphasized is that the perilymphatic space ends where the ampulla presents a wall perpendicular to the assumed direction of perilymph flow. The perilymphatic space does not end at a perpendicular wall however but in a space continuously tapering down to very small dimensions before meeting the ampullary wall as seen in Fig. 4.

From the histological pictures presented in Figs. 5 and 6 it is also apparent that the perilymphatic connective network anchors the membranous labyrinth securely to the periosteum

of the bony walls. These connections in the ampulla can always be seen in good histological sections but those around the canals are often more difficult to reproduce. Therefore it is advisable to resort to dissections like those in Figs. 5 and 6 and to use dark field microscope illumination or other suitable approaches. From these dissections it is obvious that there is very little of the ampullary wall which can face the canalicular perilymph and that the membranous ampullary walls are tightly anchored to the periosteum.

Comments on the Third Premise of Anliker and v Buskirk

These authors admit that vessels and thread-like structures are crossing the perilymphatic

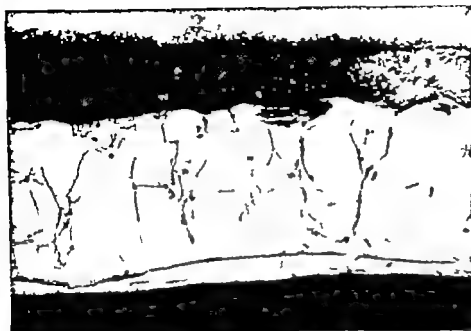


Fig. 5 Photograph of spongy network of fine connective tissue threads in perilymphatic space of the pigeon. Shown are the anterior vertical canal and part

of the ampulla. The perilymphatic tissue is clearly seen. Blood vessels and an abundance of capillaries in the perilymphatic space are also present.

space but they do not believe that these connective tissue bridges could impede the assumed motions of the perilymph. This view point would be valid if such threads were found only occasionally and in small number in the canal as a whole. We believe that Figs. 5 and 6 demonstrate that the perilymphatic space is a spongy meshwork of fine connective tissue threads. In histological sections of about 20 μ m thickness, this network can be adequately demonstrated. To assume that this meshwork offers no resistance to perilymph movement is to make an unreasonable conjecture.

Furthermore, if the assumption of cupula stimulation due to perilymph movements has any application, it should be valid for all species with a perilymphatic space. In cartilaginous fish and frogs (species used extensively in vestibular research) it is then expected that the sensitivity of the semicircular canals should be considerably lower for the following reasons. The walls of the ampullae are thick and cartilage-like. They strongly resist deformation and if indented with the point of

a needle, they slowly resume their shape due to the elasticity of the tissue. The whole labyrinth is suspended in a thick extremely viscous perilymphatic substance which gives the impression of being impossible to move by the accelerations due to head movements. When recording the action potential frequency of the ampullary nerve in these species it can be established that the sensitivity of the ampullae to acceleration is extremely high and that in principle no quantitative difference exists between these and other animal species.

Anliker & v. Buskirk (1971) criticized the results of earlier experiments to measure the rigidity of the ampullary wall (Dohlman, 1941). These experiments gave a rigidity value corresponding to the weight of 40–60 mg. Without presenting any new evidence they assume "that the membranous labyrinth could have been excessively pressurized" due to the removal of some of the perilymph over the ampulla in these experiments. This suggestion is contrary to simple physical principles. Admittedly the experiments (Dohlman, 1941) were made after

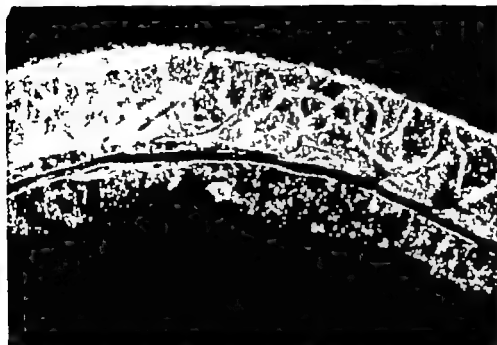


Fig 6 Photograph of spongy network of fine connective tissue threads in perilymphatic space of the pigeon. Shown are the perilymphatic tissue, blood

vessels and a tight network of capillaries covering the membranous canal wall.

removal of some perilymph covering the membranous ampullary wall. If these walls were elastic and flexible as these authors predict, removal of even a considerable part of the perilymph must necessarily result in a collapse of the unsupported membranous walls and not an increase in pressure.

Consider the following simple experiment. A rubber torus (an inner tube of a bicycle tire) is filled with water and completely submerged in a water bath as shown in Fig 7. If the tube is drawn partially out of the bath and cut

open at its vertex, air will be sucked into the tube, indicating that the internal pressure of the tube was less than that of the surrounding air. This negative pressure is due to elastic expansion of the submerged part of the rubber tube due to the pressure of the water contained in the elevated part of the tube.

In the labyrinth however the largest pressure difference between the horizontal ampulla when it is placed uppermost during the experiment, and the lowest part of the anterior vertical canal, cannot be more than at most

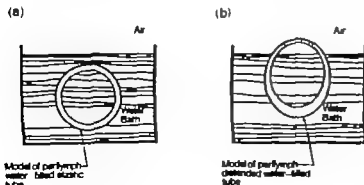


Fig 7 Model of the collapse of a water-filled torus represented by a rubber inner tube of a bicycle tire lifted out of a water bath (see text).

3 mm of water. Even if the position of ampulla and canals should be reversed, it is not easy to understand that a water pressure of 2-3 mm should be able to produce an "excessively pressurized" endolymphatic tube. Further Ewald's experiment (Ewald, 1892) involving the so-called "bridge preparation" in the pigeon, where bone and perilymph are removed from a section of the semicircular canal, always leaves an intact membranous canal. This proves that at least in this animal the walls of the canals have sufficient rigidity to withstand the pressure changes due to removal of perilymph. Further if the canal is cut, no fluid seems to escape and no air is sucked in. Therefore the statement (Dohlman 1941) that the ampulla has enough rigidity to withstand a possible impact from perilymph movement seems still to be valid.

CONCLUSION

The presence of the spongy perilymphatic tissue (Figs. 5 and 6), the angle between the canal and ampulla (Fig. 4) and the dense fibrous connections between the membranous labyrinth and the periosteum seem to be three factors which contribute to the stabilization of the membranous system and prevent any influence on cupula deflection by the perilymph. It therefore seems erroneous to consider calculations of cupula stimulation on the assumption "that the ampulla wall offers no resistance" and to disregard the influence of the perilymphatic tissue. Even if the value of the deformation resistance of the ampulla wall is inaccurate as measured by Dohlman in 1941 this resistance is not negligible and cannot be assumed to be insignificant.

In their conclusion Anliker & v Buskirk claim that they have "shown" that "it is primarily the perilymph which causes the deflection of the cupula". The premises on which these authors base their calculations, however have no factual support and are misleading as the foundation for their calculations.

A mathematical explanation for the fact

that apparently the same sensitivity to physiological stimuli can be achieved in different species in spite of the great differences in perilymphatic cross-sections would have been of greater value. The discrepancy inherent in their statement "that the contribution of the perilymph to the volumetric displacement of the cupula is almost 200 times that of the endolymph" when calculated in terms of human dimensions is apparent upon consideration of the facts that the canals of the cat have a cross-sectional perilymph area only one third that of the endolymph and that there is no evidence that the sensitivity to stimulation is different in these two species.

ZUSAMMENFASSUNG

Die Behauptung von Anliker & v Buskirk (*Acta Otolaryng* (Stockh.) 72 93), dass die Perilymphe einen kleineren Widerstand gegen Flüssigkeitsbewegungen darboten sollte als die Endolympe wegen einer Differenz in der Querschnitts-Area dieser beiden Flüssigkeits-Räume, wird zurückgewiesen. Das Netzwerk von bindegewebigen Fäden und Blutgefäßen des Perilymph-Raums durchsetzt, sowie die halbmondförmige, und nicht stützende Querschnitts-Area dieses Raumes, bietet einen beträchtlichen Widerstand der von Anliker et al. für ihre mathematische Behandlung vernachlässigt wurde. Sie haben auch eine resistenzlose perpendikuläre Wand der Ampulle angenommen, was morphologisch und physikalisch nicht mit bekannten Erfahrungen übereinstimmt. Es wird an diesen Punkten nachgewiesen, dass die Grundlagen für die Berechnungen von Anliker et al. sich auf fehlerhafte Annahmen stützt und dass nichts vorgebracht werden ist was die allgemein umfasste Ansicht von der Funktion der Endolympe für die Sinnesepithel-Reizung ändert.

REFERENCES

- Anliker M & Buskirk, W 1971 The role of perilymph in response of the semicircular canals to angular acceleration. *Acta Otolaryng* (Stockh.) 72 93
- Brener J 1874 Über die Funktion der Bogengänge des Orlabyrinthes. *Med Jahrbuch* 4 72.
- de Burlet, H 1971 Der perilymphatische Raum des Meerschweinchenohres. *Anat Anz* 53 302.
- Dohlman, G. 1941 Die Rolle der Perilymphe bei den vestibulären Reaktionen. *Arch Otorhinol* 149 5.
- Ewald, J. R. 189... Physiologische Untersuchungen über das End-Organ des Nervus octavus. *Wiesbaden, Bergmann*.
- Fernández, C. & Valentini, M. 1968. A study of

- biophysical characteristics of the cat labyrinth. *Acta Otolaryng* (Stockh.) 65: 293.
- Igarashi, M. 1965. Dimensional study of the vestibular end organ apparatus. *Second symposium on the role of the vestibular organs in space exploration*. NASA SP 115.
- Mayre, R. 1965. The match of the semicircular canals to the dynamic requirements of various species. *Second symposium on the role of the vestibular organs in space exploration*. NASA SP 115.
- Melvil Jones, G. & Speltz, K. 1963. A theoretical and comparative study of the functional dependence of the semicircular canal upon its physical dimensions. *Proc Roy Soc Ser B* 157: 403.
- Money, J., Bonen, L., Beatty, J., Kuehn, L., Sokoloff, M. & Weaver, R. 1971. Physical properties of fluids and structures of vestibular apparatus of the pigeon. *Amer J Physiol* 220: 140.
- Reijto, A. 1939. Die Rolle der Perilymphe in der Entstehung des kalorischen Nystagmus. *Acta Otolaryng* (Stockh.) 27: 270.

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DOES PERILYMPH MODIFY CUPULA DEFLECTION?

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Abstract The concept of perilymphatic actuation of the cupula proposed by Anliker & VanBuskirk (*Acta Otolaryng* 72, 93-100, 1971) has been reviewed and found that their model is based on a quite erroneous concept of the anatomy of the temporal bone. Careful study of serial slides of the temporal bone shows that the cupular deflection is caused primarily by flow of endolymph, and therefore, the mathematical model based upon a faulty view of the anatomy of the system cannot be accepted.

Since the first observation of the vestibular end-organ of pigeons and rabbits by Floarens (1824) at the beginning of the nineteenth century the vestibular apparatus has been studied morphologically in great detail, and modern anatomical and physiological investigations have added more information describing the labyrinth. It has been well accepted that cupular deflection is due to flow of endolymph and a few mathematical models have been developed for the cupula-endolymph system based upon the endolymphatic actuation of the cupula.

Recently Anliker & VanBuskirk (1971) suggested that the deflection of the cupula is caused by primarily the perilymph and they developed a new model of the perilymph-endolymph-cupula system. The concept of perilymphatic actuation of the cupula proposed by Anliker & VanBuskirk (1971) cannot go unchallenged. However sound mathematically it is unfortunately based on a quite erroneous concept of the anatomy of the vestibular end-organ. Anliker & VanBuskirk's Fig 1 which depicts an artist's drawing of their idea of the anatomy of the ampulla simply does not

exist in reality. It shows the dome of the membranous ampulla tightly up against the bony ampullary wall, suggesting that there is no perilymphatic cistern between the membranous and bony ampullary walls, and it is, therefore, impossible for perilymph to get past the membranous ampulla.

However even a superficial perusal of temporal bone sections demonstrates in both the human (Fig. 1) and the animal (Fig. 2) a substantial supra-ampullary cistern, allowing for as easy flow of perilymph past the ampulla as around the semicircular duct. Furthermore, the fibrous tissue strands supporting both the membranous semicircular duct and the dome of the ampulla could be expected both to impede perilymph flow as well as prevent deformation of the membranous ampullary wall.

In checking with Igarashi (1972) Anliker & VanBuskirk's source (Igarashi, 1969) of there being "no evidence of perilymphatic cisterns above the membranous ampulla" it is apparent that Igarashi's letter to VanBuskirk was misinterpreted.

Based on available information on the semicircular canals, cupular deflection is not caused by flow of perilymph but caused by flow of endolymph. One day we must learn how the vestibular system works and learn dynamics of the semicircular canals so that we can determine if the various mathematical models of it are sound. Meanwhile any "new model" based upon a faulty view of the anatomy or physiology of the system cannot be accepted.

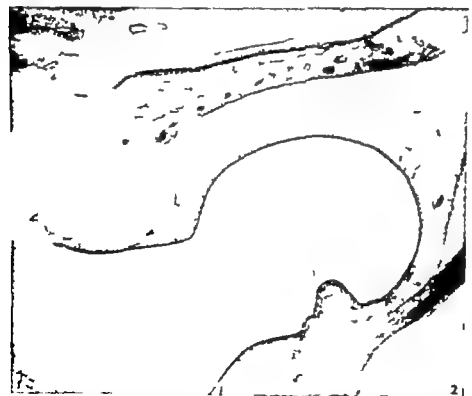


Fig 1 Human temporal bone H&E stain right center ampulla, left center utricle. The supra-ampullar and peri-ampullar cisterns are evident and rather capacious.

Fig 2 Temporal bone section of the chinchilla, H&E stain. The supra-ampullar cistern is evident in this

animal, artifact is less than in the human, due to perfusion, and the network of connective tissue strands between the membranous and bony ampullary walls are more clearly evident. The anatomic implication of this network is clear. It would be expected not only to impede perilymph flow but work against distortion of the membranous ampullary wall.

ZUSAMMENFASSUNG

Die Vorstellung, dass die Cupula durch die Perilymphe in Bewegung gesetzt wird, wie es von Anliker und VanBuskirk vorgeschlagen worden ist, ist einer näheren Prüfung unterzogen worden. Es ergab sich, dass ihr Modell auf einer gänzlich irrigen Auffassung von der Anatomie des Schläfenbeins basiert. Eine sorgfältige Untersuchung von Serienschritten des Schläfenbeins zeigt, dass die Bewegung der Cupula primär durch den Fluss der Endolymphe bewirkt wird. Das mathematische Modell, das von einer fehlerhaften Auffassung von der Anatomie des Systems ausgeht, kann somit nicht akzeptiert werden.

REFERENCES

- Anliker M & VanBuskirk, W 1971 The role of perilymph in the response of the semicircular canals to angular acceleration. *Acta Otolaryng* (Stockh.) 72 93
- Florence, P 1824 *Recherches expérimentales sur les propriétés et les fonctions du système nerveux dans les animaux vertébrés*. Crevat, Paris.
- Igarashi, M 1969 Personal Communication to VanBuskirk.
- 1972. Personal Communication to McCabe.
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NYSTAGMUS INHIBITION AS AN EFFECT OF EYE-CLOSURE

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(Received May 23 1971)

Abstract In the present investigation the nystagmus response during rotation with open eyes in darkness was compared with the response recorded during eye-closure. Eye-closure reduced the induced nystagmus response and in 20 out of 30 subjects the response decreased to such an extent that it was impossible to record by electronystagmography. A relation was found between vertical eyeball deviation during eye-closure and the degree of nystagmus inhibition. From the present investigation it appears that examinations of the peripheral vestibular function carried out while the subjects' eyes are closed often give misleading recordings which may result in incorrect conclusions. The best way of recording vestibular induced nystagmus, therefore is to test the subjects with eyes open in total darkness.

It is well known that identical vestibular stimuli may cause different nystagmus responses in different individuals, but also in the same individual on different occasions. A large number of investigations deal with the possible reasons for such variations. Some of these causes are fairly well known such as the effect of habituation (Aschan, 1954; Dodge 1923; Forsman et al. 1963; Griffith 1920; Groen, 1957; Hamersma, 1967; Hallpike & Hood, 1953; Henriksson et al., 1960; 1961; Hood & Pfaltz, 1954; Lidvall, 1961; McCabe, 1960; Sokolowski, 1963), of smoking (Tibbling, 1969; Tibbling & Henriksson, 1968), of visual fixation (Aschan, 1955; Aschan et al., 1956; Collins et al. 1961; Hallpike & Hood, 1953; Hamersma, 1967; Henriksson 1956; Mahoney et al. 1957; Sokolowski 1963; 1966; Stahle 1958) similarly the effect of variation in mental alertness (Barber & Wright, 1967; Collins, 1962; Collins & Guedry

1962; Collins et al. 1961; 1962; Gillingham, 1969; Lidvall, 1961; Mahoney et al. 1957; McLay 1962; Sokolowski, 1963, 1966).

Nystagmus inhibition is differently defined by different authors and the phenomenon is referred to in different experimental and clinical conditions. Most frequently it means that no nystagmus or only a feeble response is obtained in spite of a vestibular stimulus known to be capable of eliciting a certain response (Barber & Wright 1967; Gillingham, 1969; Groen, 1957).

The inhibition may be of a transient peripheral origin, a kind of adaptation to a repeated stimulus. It may also be of an "active" central origin, suppression considered one of the brain's methods of damping or regulating repetitive or unwanted stimuli (McCabe et al. 1964). The mechanism of this "active" suppression is still unknown however it might be caused by activity reaching the end-organ through the efferent pathways within the vestibular nerve (Dohlman et al. 1958; Gleason & Henriksson, 1964; Sala, 1965; Silverstein, 1962) reducing the peripherally elicited afferent activity at the level of the haircells, and in this way suppressing the manifestation of the peripheral vestibular stimulation. Groen (1957) is of the opinion that the inhibition takes place in the synapses of the neurons of the central vestibular system and owing to this mechanism the peripherally released impulses will not reach the oculo-motor nuclei.

There are obviously still other inhibiting

mechanisms to consider. A kind of "passive" inhibition is observed during sleep and hypnosis (Aschan et al. 1962, Johner & Perlman, 1968) and according to Barber & Wright (1967) the same phenomenon occurs in subjects who are unusually sleepy.

There is no doubt that the status of mental alertness in the subject is an important factor when the vestibular function is tested. Many authors therefore recommend some kind of mental activity during the tests to avoid nystagmus inhibition.

For the same reason visual impressions are excluded during examinations of the vestibular function to avoid inhibition by visual fixation. This exclusion is made either by eye closure or by examining the subjects in total darkness.

Using a photo-electric technique which made the subjects keep their eyes open in darkness during the test, Johnson & Torok (1970) found that the habituation effect of repeated rotatory stimulations did not take place. Torok (1970) has also shown that there is no difference in response between tests carried out during forced alertness—with mental activity—and tests made when the subject's attention is relaxed—just trying to feel comfortable and keeping the eyes open in darkness. These two investigations show that a forced state of alertness is no prerequisite for an adequate response when the examinations are carried out with open eyes in darkness. Enough mental energy seems to be provided when the subject concentrates on keeping his eyes open. The position of the eyeballs during eye-closure has been studied by many authors. About 150 years ago Bell studied the position of the eyeballs behind closed eyelids. He found that the eyeballs were elevated during eye-closure and stated that this was a physiological position of rest not only during sleep but also in conscious subjects with closed eyes in darkness.

Using a cinematographic recording of the behaviour of the eyeballs during and after eye closure, Hall (1936) discovered that the eye

balls always assumed the same position of rest in the same subject but that this position might differ from one subject to another. He found that most often in about 88% the eyeballs turned upwards and remained elevated until the eyelids opened. In some cases the eyeballs did not move at all, and in others they moved downwards. In all of the cases Finur & Eriksson (1961) found an initial elevation of the eyeballs at eye-closure which, however, was followed by a downward movement after a few seconds.

Perhaps this change of position of the eyeballs during eye-closure may affect the vestibular nystagmus. Aschan et al. (1956) however found similar responses to similar optokinetic stimuli when making recordings with the eyeballs in two different vertical positions, and concluded from this that Bell's phenomenon does not explain the differences between recordings made with the subject's eyes open or closed.

The aim of the present investigation was to find out if vestibular nystagmus elicited by angular stimulation was changed by eye-closure even though visual fixation had already been eliminated by total darkness.

METHOD

Both labyrinths were given vestibular stimulation by clockwise angular acceleration of $62^\circ/\text{sec}^2$ for 1.8 sec, followed by a constant velocity of $120^\circ/\text{sec}$. The subject was seated in the rotation chair in total darkness with his head fixed and bent forward 30° so as to place the lateral semicircular canals in the plane of rotation. The recordings were made in the horizontal plane of both eyes together and in the vertical plane of the left eye alone. By using a d.c.-technique with an infinite time constant it was possible to record the position of the eyeballs also. The recordings were made on a four-channel writer (Mingograf III Elema-Schönander Sweden) and specially devised d.c.-electrodes (Elema-Schönander Sweden) were used.

MATERIAL

The material consisted of 30 healthy people, aged 18–24 years, mostly students, 8 of whom were females and 22 males, with no history of labyrinthine disorders and no previous experience of vestibular experiments.

Performance of the test

Prior to each test the horizontal and vertical eye movements were calibrated for 20°. All tests were carried out in total darkness. Each subject was exposed to 2 different tests of 1 minute clockwise rotation with an intervening pause of 5 minutes.

1. Open eyes. the subjects were required to keep their eyes open and as far as possible to avoid blinking during the whole test.

2. Closed eyes the subjects were told to close their eyes lightly throughout the experiment and this eye-closure was initiated 3–6 seconds before the start of the rotation. Every

second test-subject was exposed to the same test but in the opposite order

Analysis of the recordings

The per rotatory nystagmus was measured and analysed by counting the frequency and by adding up the distances in millimetres from the peak of each nystagmus beat to the base line of that beat. These values were converted to eye movement degrees by means of the calibration data. Each quality was calculated in 4 periods, each of 5 sec, comprising the first 20 sec of rotation. The vertical deviation of the left eyeball during eye closure was also recorded and studied.

RESULT

Nystagmus inhibition during eye-closure

Regular and rhythmic per rotatory nystagmus was observed in all 30 subjects when recordings were made with open eyes in darkness.

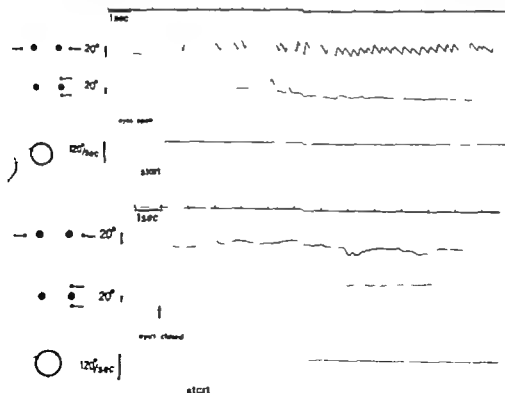


Fig. 1 Reproduction of a normal nystagmus response during rotation with open eyes in darkness (above), and in the same subject a complete inhibition of nystagmus with eyes closed during the rotation (below).

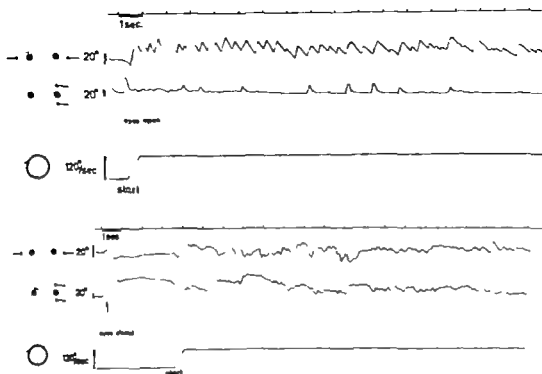


Fig. 2 Reproduction of a normal nystagmus response during rotation with open eyes in darkness (above),

and in the same subject a reduced and distorted response with eyes closed during rotation (below).

During eye-closure no nystagmus or a very reduced and distorted response was observed in 20 out of 30 subjects. The reduction and distortion in these 20 cases were so pronounced that these recordings were scored and plotted as zero. A partial reduction of the per-rotatory nystagmus was observed in 9 out of the other 10 subjects while in 1 subject there was a nystagmus response of somewhat higher intensity.

Figs. 1-3 present original d.c.-recordings of horizontal and vertical eye-movements and eye-positions before and after start of rotation with open and closed eyes in darkness, together with a recording of the angular velocity of the rotating device. As appears from the figures, regular and rhythmic responses occur during rotation with open eyes, while during eye-closure different degrees of nystagmus inhibition within different subjects can be observed.

In all subjects, diagram A in Fig. 4 shows

the mean of the total eyeball deviation in the slow component during the first 20 sec of rotation. The mean changed from 533 /20 sec with open eyes to 101 /10 sec during eye-closure. The 20 subjects with no nystagmus during eye-closure are excluded from diagram B. In spite of this exclusion a difference in the nystagmus response between rotation with open eyes and rotation during eye-closure could be observed in the resting subjects. Diagram A in Fig. 5 presents the mean frequency of beats during the first 20 sec of rotation in all subjects. The mean with open eyes was 2.5 beats/sec and during eye-closure 0.6 beats/sec. The 20 subjects with no nystagmus during eye-closure were excluded from diagram B and there was also a difference between open and closed eyes. Figs. 6 and 7 show the distribution of these calculated quantities when counted during 5-second periods. In these two figures only the 10 subjects with nystagmus during eye-closure are plotted.

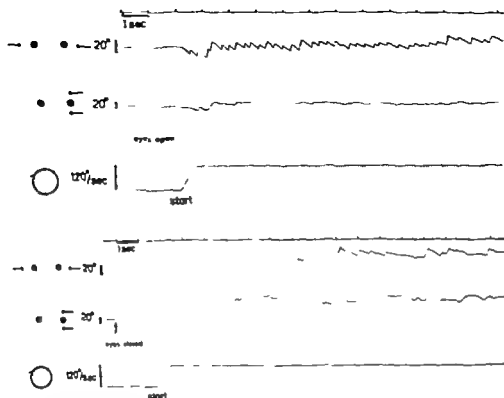


Fig. 3 Reproduction of a normal nystagmus response during rotation with open eyes in darkness (above), and in the same subject a partial inhibition of the nys-

tagmus response when the eyes are closed during rotation (below).

Position of eyeballs during eye-closure

In all subjects there was an initial elevation of the eyeballs at eye-closure. From this initial elevation the eyeballs turned down again behind the eyelids in some of the cases, while in others the eyeballs remained elevated as long as the subjects were told to keep their eyes closed. The speed and degree of the downward movement varied from one subject to another. Repeated tests showed the same patterns in the same subjects though the patterns could be slightly or moderately modified. Fig. 8 shows examples of different patterns of eye-balls positions during eye-closure.

Relation between vertical eyeball deviation during eye-closure and nystagmus inhibition

In the recordings with total nystagmus inhibition during eye-closure the eyeballs remained elevated behind the eyelids as long as the sub-

jects were told to keep the eyes closed (Fig. 1). From the recordings with partial inhibition of nystagmus during eye-closure it appeared that the eyeballs turned down behind the eyelids after the initial elevation (Fig. 3). Fig. 9 shows that different positions of the eyeballs during eye-closure are combined with different degrees of nystagmus inhibition. In Fig. 9a the eyeballs remain elevated during eye-closure and the induced nystagmus is completely inhibited during this period. The nystagmus is not released until the subject is told to open his eyes, at which moment the eyeballs turn down again. Fig. 9b shows an initial brief eyeball movement upwards at eye-closure and a simultaneous nystagmus inhibition during this very period. The eyeballs then turn down behind the eyelids and the nystagmus is released again though the eyelids are still closed.

A statistical evaluation has been performed using Student's *t*-test. The mean frequency

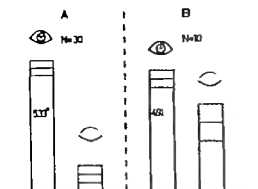


Fig 4 Total of eye-deviation in slow components during first 20 seconds of rotation with open and closed eyes in darkness. Results are given as mean \pm S.E.M. (). Diagram A gives the results with all 30 subjects included. Diagram B gives the results when the 20 subjects with no nystagmus during eye-closure are excluded.

during the first 20 seconds of rotation was significantly higher with open eyes than with closed eyes ($p < 0.001^{***}$). The mean total eye-deviation in the slow components was also significantly higher with open eyes than during eye-closure ($p < 0.005^{**}$). The statistical evaluation was performed with the mean of all subjects examined with open eyes and the mean of the 10 subjects with nystagmus during eye-closure.

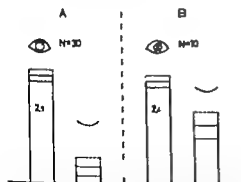


Fig 5 Frequency of nystagmus beats during rotation with open and closed eyes in darkness, counted during first 20 sec of rotation. Results are given as mean \pm S.E.M. Diagram A gives the results with all 30 subjects included. Diagram B gives the results when the 20 subjects with no nystagmus during eye-closure are excluded.

DISCUSSION

Nystagmus inhibition as an effect of eye-closure

As this study has shown eye-closure may reduce induced nystagmus response and some times even inhibit the response completely. On the other hand, many authors have found that eye-closure facilitates the induced nystagmus response and prolongs duration of nystagmus (Aschan et al., 1956; Hamersma, 1967; Sokolowski, 1963; Stahle, 1958). Though these different results seem irreconcilable there is no real contradiction. In the first case, the nystagmus response obtained with open eyes is compared with the response obtained during eye-closure, and both tests are carried out in darkness. In the second case the comparison is between open and closed eyes in light and the difference found in these tests must be ascribed to visual fixation. As visual fixation in the present study is already eliminated with darkness, there is no comparison between visual fixation and non-visual-fixation but between open and closed eyes.

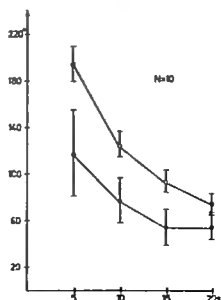


Fig 6 Total of eye-deviation in slow components with open (O), and closed (●) eyes in darkness counted during 5 sec periods from start of rotation. Results are given as mean \pm S.E.M. Only the 10 subjects with nystagmus during eye-closure are plotted.

ing eye-closure and nystagmus inhibition. A more pronounced inhibition occurred when the eyeballs remained elevated during eye closure than in cases where the eyeballs turned down again after the initial elevation. It was also seen how the nystagmus beats returned or started when the eyeballs turned down from the elevated position. In this case it was necessary to test the possibility that the change in the electrical field projection was responsible for the change of the recorded nystagmus response. This was checked by calibration experiments with the head bent forwards 30° and backwards 40° while the subject was made to make the same calibration movements with his eyes. Bending of the head in this way still allowed fixation of the calibration target. In this way the subject's eyes made the same horizontal deviation though with the eyeballs in a different vertical position. All of these calibrations, however, gave the same deflection of the tracing. From this it can be concluded that the changes in the electrical field projection is not responsible for the change of the recorded nystagmus response.

Changes in the activity inhibition or activation of the different extra-ocular muscles at eye-closure due to the reciprocal innervation may be in conflict with the vestibularly induced activity and thus cause nystagmus inhibition.

It is observed how the tonic activity of the extra-ocular muscles is inhibited during sleep (Björk & Kugelberg, 1953; Breinin & Moldaver 1955), and it is also known that vestibular stimulation during sleep causes no nystagmus response. As the proprioceptive system of the body also includes the extra-ocular muscles (Breinin 1957), it might be assumed that a stimulation of the muscles turning the eyeballs upwards might cause a change of tonus in the other extra-ocular muscles. This might also be in conflict with the vestibularly induced eye movements. The reciprocal innervation causing the elevation of the eyeballs at eye-closure also changes the mechan-

ical prerequisites for the eyeballs movements in the horizontal plane. When the eyeballs are elevated the relation between the insertion of the eye-muscles and the axis of rotation is changed and perhaps this might be one of the factors contributing to the variations.

CONCLUSION

This investigation shows that recording of induced nystagmus during eye-closure often gives reduced and distorted responses which might lead to incorrect conclusions about the peripheral vestibular function. It is well known that there is a considerable variation of the nystagmus output owing to different experimental conditions. The present investigation has shown that these variations are more pronounced when the tests are made with the eyes of the subject closed. As the eye muscles seem to have no position of rest (Björk, 1954; Breinin & Moldaver 1955), it seems to be in accordance with physiological conditions to investigate vestibular induced eye-movements with the eyes open in darkness when only the vestibular induced activity affects the movements of the eyeballs. This is preferable to examining the subjects during eye-closure when both the activities evoked by the reciprocal innervation and the proprioceptive system might be in conflict with the vestibular induced activity. It may seem impractical to record nystagmus in total darkness, but in our laboratory the technical equipments are arranged so as to make it possible to work from outside the dark room. This arrangement seems to be well worth the trouble and the expense.

ZUSAMMENFASSUNG

Durch Winkelstimulation ausgelöster Nystagmus wurde an 30 gesunden Versuchspersonen studiert. Die durch Drehbewegungen ausgelöste Nystagmusreaktion wurde an Personen mit geöffneten Augen mit derjenigen bei geschlossenen Augen verglichen. Stimulierende Versuche wurden in vollkommener Dunkelheit durchgeführt. Bei Lidchluss verringerte sich der induzierte Nystagmus, so dass bei 70% von 30 Versuch-

personen keine Reaktion mehr elektromyographisch festgestellt werden konnte. Möglicherweise besteht ein Zusammenhang zwischen einer Aufwärtsbewegung des Auges bei Lidchluss und dem Aussetzen der Nystagmusverringernng. Schlussfolgerung, dass eine Fehlerquelle bei der elektromyographischen Aufzeichnung von durch Drehbeschleunigung ausgelöstem Nystagmus beobachtet werden kann, wenn die untersuchte Person während des Versuches in vollkommener Dunkelheit die Augen offen hält anstatt sie zu schließen. Aus der vorliegenden Untersuchung geht hervor dass ENG-Aufzeichnungen mit geschlossenen Augen oft zu irreführenden Resultaten führen, die falsche Rückschlüsse betreffs der peripheren vestibulären Funktion zur Folge haben können.

REFERENCES

- Adams, A. 1957 Nystagmographische Untersuchungen über den Lidnystagmus und die physiologische Koordination von Lidschlag und rascher Nystagmusphase. *Arch Oth. der Kehlkopfheilk.* 170 543
- Aschan, G. 1954 Response to rotatory stimuli in fighter pilots. *Acta Otolaryng* (Stockh.), Suppl. 116 4
- 1955 The caloric test. *Acta Soc Med Upsal* 60 99
- Aschan, G. Bergstedt, M. & Stahle, J. 1956. Nystagmography. *Acta Otolaryng* (Stockh.), Suppl. 129 1
- Aschan, G. Finer, B. L. & Hagbarth, K. E. 1962. The influence of hypnotic suggestion on vestibular nystagmus. *Acta Otolaryng* (Stockh.) 55 97
- Barber, H. O. & Wright, B. A. 1967 Release of nystagmus suppression in clinical electronystagmography. *Laryngoscope* 77 1016
- Byrjork, A. 1954 Electromyographic studies on the coordination of antagonistic muscles in cases of abdomens and facial palsy. *Brit J Ophthalmol* 38 605
- Byrjork, A. & Kugelberg, E. 1953 The electrical activity of the muscles of the eye and eyelids in various positions and during movement. *Electroenceph. Clin. Neurophysiol* 5 595
- Breinin, G. M. 1957 Electromyographic evidence for ocular muscle proprioception in man. *Arch Ophthalmol* (Chic.) 57 176
- Breinin, G. M. & Moldaver, J. 1955 Electromyography of the human extraocular muscles. *Arch Ophthalmol* (Chic.) 54 200
- Collins, W. E. 1962. Effects of mental act upon vestibular nystagmus. *J Exp Psychol* 63 191
- Collins, W. E., Crumpton, G. H. & Posner, J. B. 1961 Effects of mental activity on vestibular nystagmus and the electroencephalography. *Nature* 190 194
- Collins, W. E. & Guedry, F. E. 1962. Arousal effects and nystagmus during prolonged constant angular acceleration. *Acta Otolaryng* (Stockh.) 54 349
- Collins, W. E., Guedry, F. E. & Posner, J. B. 1962. Control of caloric nystagmus by manipulating arousal and visual fixation distance. *Ann Otol* 71 187
- Dodge, R. 1923 Habituation to rotation. *J Exp Psychol* 6 1
- Dohlman, G. Farfashky, J. & Salonna, F. 1958. Centrifugal nerve-fibres to the sensory epithelium of the vestibular labyrinth. *J Laryng* 72 984
- Flour, E. & Erlanson, L. 1961 Nystagmographic recording of vertical eye movements. *Acta Otolaryng* (Stockh.) 53 486
- Forssman, B., Hennikson, N. G. & Dolowitz, H. A. 1963 Studies on habituation of vestibular reflexes. VI. Habituation in darkness of calorically induced nystagmus, lateroocclusion and vertigo in man. *Acta Otolaryng* (Stockh.) 56 663
- Gillingham, K. K. 1969 Mental arithmetic during electronystagmographic testing. *Ann Otol* 78 575
- Gleimser, L. & Hennikson, N. G. 1964 Efferent and afferent activity pattern in the vestibular nerve of the frog. *Acta Otolaryng* (Stockh.), Suppl. 192 90
- Griffith, C. R. 1920 The organic effects of repeated bodily rotation. *J Exp Psychol* 3 15
- Gross, J. J. 1957 Adaptation. *Pract Otorhinolaryng* (Basel) 19 524
- 1957 The semicircular canal system of the organs of equilibrium. *Physiol Med Biol* 1 225
- Hall, A. J. 1936. Some observations on the acts of closing and opening the eyes. *Brit J Ophthalmol* 20 257
- Hallpike, C. S. & Hood, J. D. 1953 Fatigue and adaptation of the cupular mechanism of the human horizontal semicircular canal. *Proc Roy Soc Med* 46 542
- Hamerusa, H. 1967 *The caloric test. A nystagmographic study* Pp. 1-1, Lutz, Berger op Zoom, Holland.
- Hennikson, N. G. 1956 Speed of slow component and duration in caloric nystagmus. *Acta Otolaryng* (Stockh.), Suppl. 125 156
- Hennikson, N. G., Fernández, C. & Kohut, R. 1960 The caloric test in the cat. *Acta Otolaryng* (Stockh.) 53 21
- Hennikson, N. G., Kohut, R. & Fernández, C. 1961 Studies on habituation of vestibular reflexes. *Acta Otolaryng* (Stockh.) 53 333
- Hood, J. D. & Pfaltz, C. R. 1954 Observations upon the effects of repeated stimulations upon rotational and caloric nystagmus. *J Physiol* 124 130
- Jobner, C. H. & Portman, H. H. 1968 Hypnosis and vestibular function. *Ann Otol* 77 126
- Johnson, D. D. & Torok, H. 1970. Habituation of nystagmus and emulsion of motion after rotation. *Acta Otolaryng* (Stockh.) 69 206
- Lidvall, H. F. 1961 Vertigo and nystagmus responses to caloric stimuli repeated at short intervals. *Acta Otolaryng* (Stockh.) 53 33
- 1961. Vertigo and nystagmus responses to caloric stimuli repeated at short and long intervals. *Acta Otolaryng* (Stockh.) 53 507
- Mahoney, J. L., Harlan, W. L. & Backford, R. G. 1957 Visual and other factors influencing caloric

- nystagmus in normal subjects. *Arch Otolaryng* (Chic.) 66 46
- McCabe B. F. 1960. Vestibular suppression in figure skaters. *Trans Amer Acad Ophthalmol Otolaryng* 64 64
- McCabe, III F., Gillingham, K. & Arbor A. 1964. The mechanism of vestibular suppression. *Ann Otol* 73 816.
- McLay K. 1964. An assessment of methods of vestibular testing in clinical practice. *J Laryng* 76 34
- Nathanson, M., Bergman, P. S. & Anderson, F. J. 1957. Significance of oculoccephalic and caloric responses in the unconscious patient. *Neurology* 7 829
- Sala, O. 1965. The efferent vestibular system. Electrophysiological research. *Acta Otolaryng* (Stockh.), Suppl. 197
- Silverstein, H. 1962. The cortical influence on the vestibular apparatus. *Arch Otolaryng* (Chic.) 76 158
- Stable J. 1958. Electronystagmography in the caloric and rotatory tests. *Acta Otolaryng* (Stockh.), Suppl. 137
- Sokolowski, A. 1963. Factors influencing nystagmus due to rotation in normal subjects. *J Laryng* 77 185
- 1966. The influence of mental activity and visual situation upon caloric induced nystagmus in normal subjects. *Acta Otolaryng* (Stockh.), 61 209
- Tibbling, L. 1969. The influence of tobacco smoking, nicotine, CO and CO on vestibular nystagmus. *Acta Otolaryng* (Stockh.) 68 118
- Tibbling, L. & Henriksson, N. O. 1968. Effect of cigarette smoking on the vestibular nystagmus pattern. *Acta Otolaryng* (Stockh.) 65 518
- Torok, N. 1970. The effects of arousal upon vestibular nystagmus. *Advance Otorhinolaryng* 17 76.
- Uemura, T. 1967. Electronystagmography and its clinical significance. *Otologie Fukuoka* 13 Suppl. I 67

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CENTRAL MASKING IN NORMAL LISTENERS

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Abstract. The central masking phenomenon (CM) was studied with a clinical audiometric paradigm in a group of 40 normal-hearing subjects. Contralateral maskers of wide-band noise and narrow-band noise were used each at 40 dB SL to evaluate thresholds for 500, 1000, 2000 and 4000 Hz tones. CM was shown to increase in dB with increase in frequency of the test tone and with decrease in masker bandwidth. A corrective factor of 5 dB to 10 dB was suggested to account for the effect in clinical audiometric evaluation.

Central masking (CM) is a recognized phenomenon demonstrated by a shift of auditory threshold in one ear upon presentation of a contralateral masker. That is, increased intensity is necessary to maintain threshold in the test ear when a contralateral masker is at a level which is not intense enough to directly shift the threshold of the test ear by transcranial stimulation. Various investigators have demonstrated that the true CM effect is not due to transcranial physical stimulation of the test ear nor to activation of the bilateral acoustic reflex. Therefore, due to lack of evidence regarding these strictly peripherally based phenomena, it has been hypothesized that the effect is somehow mediated by central auditory pathways with or without possible efferent fiber participation (Ingham, 1957 and 1959; Lidén et al., 1959).

Wegel & Lane (1924) provided the first modern description of the phenomenon and called it "central masking". Ingham's more

recent work provided the basis for contemporary investigations of CM under the paradigm of psychoacoustic experimentation. Zwislöcki and his associates (1965, 1966, 1968) and Dirks and his associates (1964, 1965, 1966) have extended the knowledge of CM in two series of experiments evaluating various parameters including frequency, intensity and temporal aspects of stimulus versus masker designs. Their work was with pulsed and continuous stimuli and maskers under conditions not usually encountered in traditional clinical pure tone threshold evaluation. Their findings suggested that the CM effect (in dB) increases with increased intensity of the masker that it has its maximum effect upon initial introduction of the masker with quick reduction to a relatively stable shift after about 200 msec onset, that the shift decreases as the spectrum level of the masker is broadened around the test tone and that it is statistically stable and repeatable.

The use of masking in one ear is necessary in the clinical evaluation of threshold whenever some critical difference is shown between the two ears so as to prevent a "shadow" response from the better ear. A conventional point of view suggests the need for masking in the better ear for air-conduction thresholds whenever the bone-conduction threshold for that ear is about 50 dB better than the air-conduction threshold for the poorer (test) ear. The need for contralateral masking more often is required in bone-conduction measurements

This work was supported in part by a training grant, No. 1T1NB05553 from the National Institutes of Health.

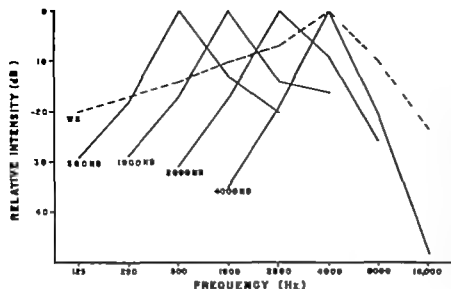


Fig 1 Acoustic spectra of wide-band (WB) and four narrow-bands (NB) of white noise. Each curve represents the noise at an overall 80 dB SPL. Identical relative curves (within \pm dB) were produced at 60 dB overall SPL.

where interaural attenuation may be as low as 0 dB

The investigation of CM as a phenomenon in the clinical evaluation of hearing has received minimal scientific attention. A number of authors have suggested a general figure for the threshold shift. Palva (1954) characterizes the change in threshold as "generally less than 10 dB". Liden et al. (1959) state that the effect is 5 to 15 dB remaining relatively constant as the masker level increases. An approximate 5 dB CM figure also has been suggested by Studebaker (1964), Price (1971) and Martin (1972). These reports were in terms of general clinical observations without formal investigation.

The experiment reported here was completed to determine the CM effects in normal listeners under the paradigm of clinical audiometric techniques, stimuli and maskers. Effects of test frequency and masker spectrum were evaluated statistically as an initial step toward adequate description of the phenomenon as a guide for the clinician.

EXPERIMENTAL DESIGN

Frequencies chosen for evaluation were 500, 1000, 2000 and 4000 Hz. These represented

a range appropriate for clinical as well as experimental, use without introducing problems of tactual stimulation (from lower frequencies) or problems of measurement reliability and validity (from higher frequencies).

Experimental conditions included measurement without contralateral masking and with wide-band (WB) white noise and narrow-band (NB) white noise contralateral maskers. These types of maskers are both in general clinical use.

A 40 dB sensation level (SL) contralateral masker was used. This level is often required to mask out a normal contralateral ear in clinical audiometry. Further, although this level might produce CM (as desired in this study) it was not great enough to mask the tone in the contralateral (test) ear by transcranial stimulation (Saunders & Rintelman, 1964; Dirks & Malmquist, 1965; Dirks & Norris, 1966; Studebaker, 1967; Shimizu, 1969).

The design provided different entry points to a consistent serial order of presentation for four frequencies by three conditions. A different entry point was drawn blindly for groups of three to four subjects. This method was used to help preclude learning practice and some order effects.

METHOD

Subjects

Forty normal-hearing adults served as subjects for the experiment. All subjects had normal hearing bilaterally (defined as -5 dB hearing level (HL) through 15 dB HL, ISO 1964) at octave intervals for the frequency range 250 Hz through 8 000 Hz. They were between the ages of 19 and 39 with a mean age of 27.1 years. All subjects were unpaid volunteers who were experienced in taking hearing tests.

No subject was used who had a -10 dB HL due to the fact that this was the limit of the audiometer and even better hearing was possible. An additional criterion was that no subject showed greater than a 10 dB difference between threshold for the two ears at any frequency. This choice was made partly due to requirements for a subsequent experiment which was performed but is not reported here.

Test environment and equipment

Testing was completed in an Industrial Acoustics test suite, Model 1600 ACT. The basic equipment used for the study was an Allison Model 22 audiometer. The signals were presented through TDH 39 earphones mounted in MX/AR cushions. The calibration and linearity of the audiometer was checked and monitored appropriately with a Brüel and Kjær sound level meter.

The audiometer served as the source for the pure-tone stimuli and the WB noise masker. The NB masking signal was supplied by the Allison Model 26 NB masking filter attachment for the Model 22 audiometer. Octave band spectral analysis of the two types of white noise were determined with the Brüel and Kjær equipment. The acoustic spectra are shown in Fig. 1.

Determination of threshold sensitivity

Air-conduction threshold measurements were made in the following frequency order: 1 000, 2 000, 4 000, 8 000 recheck of 1 000, 500, and 250 Hz. Intensity levels were varied in

steps of 5 dB. Presentation of the tone was for a duration of about one to two seconds. The tone was first presented at 40 dB HL. When the subject signaled that the tone was heard, the examiner then attenuated it to a -10 dB HL and ascended in 5 dB steps until the subject again signaled that the tone was heard. The latter procedure was repeated until the lowest intensity level was established at which the subject heard the tone two out of four presentations. This point was designated as threshold.

Threshold measurements for the masking signals were made immediately following the establishment of the pure-tone thresholds. If the two ears were exactly the same for the pure-tone measurements, the right ear arbitrarily was selected for the tone stimulus and the masker thresholds were made only for the left ear. In cases where there was a 10 dB or less difference between ears, the speech frequencies were averaged and the better ear was used for the masker presentation. This precaution was made to reduce the possibility of the masker affecting the peripheral mechanism of the test ear by cross-hearing.

Masker threshold measurements were made in the following order: WB white noise and then NB white noise for 1 000, 2 000, 4 000 and 500 Hz bands. Sensitivity evaluation was made in 5 dB steps using the ascending technique and the 50% criterion as previously described.

Evaluation of CM

Prior to the evaluation of threshold for each frequency and each time it was tested, the subject heard a reference tone of that frequency at about 20 dB SL. Subjects were instructed to disregard the noise when the threshold was established in the presence of the contralateral maskers. The following order was used during the experimental procedure: (1) the subject was presented with the reference tone for the frequency, (2) the reference tone was terminated, (3) the masker was presented on and about two seconds later.

Table I Amount of central masking (in dB) produced under two experimental conditions

Wide-band (WB) and narrow-band (NB) maskers are shown according to sensation level

	Contralateral masking	Test frequency (in Hz)			
		500	1 000	2 000	4 000
Mean	40 dB WB	1.8	2.8	3.0	4.6
	40 dB NB	3.5	3.5	4.5	6.6
S.D.	40 dB WB	2.7	3.2	3.2	3.5
	40 dB NB	3.1	2.8	3.3	3.2
Range	40 dB WB	0-10	0-10	0-10	0-10
	40 dB NB	0-10	0-10	0-10	0-15

was presented at the previously determined unmasked threshold for about one or two seconds. If there were no response the tone and masker were terminated a 20 second rest period followed and the procedure was repeated with the tone presentation at the next higher 5 dB SL. Each NB masker was used only in the evaluation of its comparable single test frequency CM was determined by comparing the unmasked and masked thresholds.

RESULT

Date from which results are drawn are present in Table I

Frequency effects

Two significant results merit attention. First, appreciable amounts of CM were shown for all frequencies under at least one experimental condition in terms of mean CM. Second, 10 dB or greater CM was shown for all frequencies under all conditions by one or more subjects that is, in terms of individual test results.

Individual Wilcoxon tests (Siegel 1956) were performed in order to determine if there were significant differences for all frequency comparisons under each experimental condition. Table II indicates that as the difference in Hz between test frequencies became greater the obtained CM difference for those frequencies tended to become significant. This tendency

Table II Difference scores (Wilcoxon matched-pairs signed ranks test) for central masking according to frequency of test tones for each masker. Maskers are shown in SL for WB and NB conditions

Test frequency (in Hz)	Contralateral masker	
	40 dB WB	40 dB NB
500 vs. 1 000	1.50	0
500 vs. 2 000	2.13	0.90
500 vs. 4 000	3.30	3.65
1 000 vs. 2 000	0.44	1.18
1 000 vs. 4 000	2.16	3.49
2 000 vs. 4 000	1.93	2.82 *

Significant at 1% level.

Significant at 5% level.

was true under every condition. For example, the CM values for 500 Hz and 4 000 Hz always displayed significantly different rankings at the 0.01 level of confidence. In contrast, the CM differences between 500 Hz and 1 000 Hz were not of significance. Similar results were shown for all other frequency comparisons.

Masker effects

Wilcoxon test results for masker effects are shown in Table III. Results indicate that as the band width of the masker narrowed, the CM effect tended to become significantly greater.

DISCUSSION

The findings of this investigation are in general accord with the results of psychoacoustic studies of CM. Occurrence of 5 dB or greater CM was greater under this clinical design than in studies previously reported.

Table III Difference scores (Wilcoxon matched-pairs signed ranks test) for central masking according to masker conditions for each frequency

Contralateral maskers (in dB SL)	Test frequency (in Hz)			
	500	1 000	2 000	4 000
40 WB vs. 40 NB	-0.89	1.4	-1.4	-6.6

Significant at 1% level.

Ingham (1959) and Dicks & Malmquist (1965) reported general increase in CM with increase in the test tone frequency. However both studies used only pure-tone maskers or a single NB masker for all test tones evaluated. Sherrick (1961) found slight, but regular increase of CM with increased test frequency but under four sets of pulsed tone stimuli which are not comparable to the design of the present study. Masker effects upon CM also generally follow results of previous psycho-acoustic investigators. Results are comparable to those obtained by Ingham (1957), Studebaker (1962), Dirks and associates (1964, 1965, 1966) and Zwillocki et al. (1968) but each of these studies used only one type of masker and/or a single test tone under conditions not directly comparable to clinical techniques.

Clinical paradigm investigations of CM for frontal bone-conduction thresholds have been reported by Naunton (1957) and Studebaker (1962). The designs and methods of the two experiments do not allow direct comparison with this study. Other rather anecdotal-type remarks of CM in clinical threshold study have been expressed by Dirks (1964), Martin et al. (1965), Price (1971), and Martin (1972). They generally advocate subtraction of about 5 dB threshold to account for CM when contralateral masking is used.

CM results from this study were separated by degree according to frequency and masker condition. Table IV shows the percentage of subjects exhibiting 0 dB, 5 dB or greater CM, and 10 dB or greater CM. In seven of the eight experimental tasks performed by each subject, 50% or more of the subjects showed 5 dB or greater CM. Findings suggest that if one chooses not to subtract for CM, one may overestimate the degree of loss. In cases where masking is not necessary for air-conduction measures, but is necessary for bone-conduction thresholds, the amount of an air-bone gap shown may be underestimated.

General conclusions suggest that it is appropriate to subtract at least 5 dB to account for CM, particularly when testing at fre-

Table IV. Percentage of subjects exhibiting three categories of central masking according to SL and type of contralateral masking

WB = wide-band. NB = narrow-band

Test frequency	Central masking (in dB)		
	0 (%)	5+ (%)	10+ (%)
500 Hz 40 dB WB	67.5	32.5	2.5
40 dB NB	40	60	10
1000 Hz 40 dB WB	50	50	7.5
40 dB NB	35	65	5
2000 Hz 40 dB WB	47.5	52.5	7.5
40 dB NB	30	70	15
4000 Hz 40 dB WB	27.5	72.5	20
40 dB NB	5	95	30

quencies above 1000 Hz. The use of an NB masker has gained considerable clinical support in recent years due to its increased effectiveness relative to comparable WB maskers and due to the ease with which it is accepted by the patient. These results temper the acceptance of an NM masker due to its "enhancement" of CM. A rather large proportion of the subjects showed CM of 10 dB or greater with the NB masker. One may sacrifice threshold validity to some degree by using an NB masker which enhances CM by 10 dB to 15 dB.

Ingham (1957, 1959) skillfully has summarized possibilities of peripheral and central origins of the phenomenon. It is obvious that the low-level masker used here could not contribute to peripheral causes of CM by cross-hearing nor through contractions of the stapedius muscle. A process involving simple distraction, factors of decreased motivation or indecisiveness under masking conditions also might be hypothesized to account for CM. In the design of this study order practice and learning effects were controlled. The highly significant frequency-related findings, therefore, would not seem to be related to mental factors which would be expected as relatively equally distributed among all frequencies.

ZUSAMMENFASSUNG

Das Phänomen zentraler Maskierung (CM) wurde an einer Gruppe von 40 Personen mit normalem Gehör mit Hilfe einer klinischen audiometrischen Methode studiert. Kontralaterale Maskierung durch hoch- und niederfrequenziges Geräusch wurde bei 40 dB SL benutzt, um die Schwelle für Töne von 500, 1000, 2000 und 4000 Hz zu bestimmen. Es wurde gezeigt, dass die CM in dB zunahm, wenn der Testton an Frequenz zunahm und die Bandbreite der Maskierung abnahm. Ein Korrekturfaktor von 5 dB bis 10 dB wurde vorgeschlagen, um den Effekt der klinischen audiometrischen B-Stimmung auszugleichen.

REFERENCES

- Dix, D. 1964 Bone-conduction measurements. *Arch Otolaryng* (Chic.) 79 594
- Dix, D. & Malmquist, C. 1964 Changes in bone-conduction thresholds produced by masking in the non-test ear. *J Speech Hearing Res* 7 271
- 1965 Shifts in air conduction thresholds produced by pulsed and continuous contralateral masking. *J Acoust Soc Amer* 37 631
- Dix, D. & Norris, J. C. 1966 Shifts in auditory thresholds produced by ipsilateral and contralateral maskers at low-intensity levels. *J Acoust Soc Amer* 40 1
- Ingham, J. E. 1957 The effect of monaural sensitivity of continuous stimulation of the opposite ear. *Quart J Exp Phys* 9 3
- 1959 Variations in cross-masking with frequency. *J Exp Psychol* 58 199
- Likén, G., Nilsson, G. & Anderson, H. 1959 Masking in clinical audiometry. *Acta Otolaryng* (Stockh.) 50 1-5
- Martin, F. N. 1971. *Clinical audiology and masking*. Bobbs-Merrill, Indianapolis.
- Martin, F. N., Bailey, H. A. T. Jr & Pappas, J. J. 1965 The effect of central masking on threshold for speech. *J Audiol Res* 5 293
- Naughton, R. F. 1957 Clinical bone-conduction audiometry. *Arch Otolaryng* (Chic.) 66 281
- Palva, T. 1954 Masking in audiometry. *Acta Otolaryng* (Stockh.), Suppl. 118 156
- Price, L. L. 1971 Pure tone audiometry. In *Audiological assessment* (ed. E. Rose), p. 167. Prentice-Hall, Englewood Cliffs, N.J.
- Sanderson, J. W. & Rintelman, W. F. 1964 Masking in audiometry. *Arch Otolaryng* (Chic.) 80 541
- Sherrick, C. E. & Mangabeira-Albermar, P. L. 1951 Auditory threshold shifts produced by simultaneous pulsed contralateral stimuli. *J Acoust Soc Amer* 23 1381
- Shimizu, H. 1969 Influence of contralateral noise stimulation on tone decay and SISI tests. *Laryngoscope* 79 155
- Siegel, M. 1956 *Nonparametric statistics for the behavioral sciences*. McGraw-Hill, New York.
- Studebaker, G. A. 1962. On masking in bone-conduction testing. *J Speech Hearing Res* 5 15
- 1964 Clinical masking of air and bone-conduction stimuli. *J Speech Hearing Dis* 29 23
- 1967 Clinical masking of the non-test ear. *J Speech Hearing Dis* 32 360.
- Wegel, R. L. & Lane, C. E. 1924 The auditory masking of one pure tone by another and its probable relation to the dynamics of the larynx. *Physical Res* 23 766.
- Zwischki, J., Brining, E. & Glanz, J. 1965 Three time parameters of central masking (abstract). *J Acoust Soc Amer* 38 938
- 1968 Frequency distribution of central masking. *J Acoust Soc Amer* 43 1267
- Zwischki, J. & Johnson, E. 1966. Central masking on time and intensity interaction (abstract). *J Acoust Soc Amer* 40 1251
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DIRECTIONAL AUDIOMETRY

VII. *The Influence of Azimuth on the Perception of Speech in Aided and Unaided Patients with Binaural Hearing Loss*

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(Received May 15 1972)

Abstract Twenty patients with binaural hearing loss varying from a Pure Tone Average (PTA) of 28 dB hearing level to 45 dB hearing level were examined. The Directional Threshold of Intelligibility (DTI) was measured both without and with background noise, without and with hearing aids. Without background noise, the DTI with binaural hearing aids was found to be significantly better than the DTI with monaural hearing aid in 3 of the 4 positions of the signal loudspeaker. With background noise, the DTI with binaural hearing aids was found to be significantly better than with monaural hearing aid in 6 of the 16 combinations of signal and noise loudspeakers. For all the 70 experimental listening conditions with background noise we found a significantly better DTI with binaural hearing aids than with monaural in 11 of the 20 patients. For the 16 experimental listening conditions with background noise we found a significantly better DTI with binaural hearing aids than with monaural in 13 of the 20 patients. The results indicate that binaural hearing aids are to be preferred.

As early as 1912 Soret took out a patent for headborne stereophonic hearing aids (Hirsh 1950). However although treatment by means of stereophonic hearing aids is not new there is still disagreement as to whether monaural or binaural hearing aids should be used. Many attempts have been made to throw light on this matter but the experimental conditions under which the various investigators have worked differ so that their results cannot be directly compared.

Watson (1942), Bender & Wilg (1960) Kodman (1961), Whettnall (1964), Bentzen et al.

(1965), and Jordan et al. (1967) all concluded from their experience that the advantage lay with binaural hearing aids.

Dirks & Carhart (1962) stated that binaural hearing aids seemed to be better than monaural, but they did not find the difference very pronounced. Carhart (1958) demonstrated the advantages of binaural treatment but was also aware of the difficulties involved in such treatment. Haskins & Hardy (1960) were positive about binaural hearing aids but also mentioned that patients had refused to use them. Hedgecock & Sheets (1958), Markle & Aber (1958), Wright & Carhart (1960), and Di Carlo & Brown (1960) did not perceive any dramatic difference between the effect of binaural and monaural hearing aids. However it should be mentioned that in Hedgecock & Sheets (1958) experimental condition, speech and noise came from the same loudspeaker so that their investigations would correspond more to experiments made with hearing aid with Y-cord.

Jerger & Dirks (1961) and Jerger et al. (1961) did not find binaural hearing aids better than monaural. In their experiments the head was allowed free movement with the result that the experimental subject could move the head into the most favourable position. The deaf do almost as a reflex

Belzile & Markle (1959) observed that in patients with conductive and with perceptive hearing loss, binaural hearing aids were better than monaural. For both groups 50% discrimination could be achieved in the presence of 10 dB more noise while wearing binaural hearing aids than could be achieved while wearing monaural hearing aid.

Since 1964 the State Hearing Center in Aarhus has used as routine to treat patients with double-sided hearing loss with binaural aids, and the clinical experience has been good. In this paper an attempt will be made to throw some light on monaural and binaural treatment with headborne apparatus by comparing the two methods of treatment.

METHOD

The apparatus and method has been described in detail earlier (Tønning 1970 1971 *a* and *b* 1972 *a* *b* and *c*).

Throughout the following, the dB reference level is 0.0002 dyne per square centimetre. Hearing loss in dB refers to British Standard 2497 1954. The relationship between British Standard and ISO Standard is shown in Table I (See also Whittle & Delany 1966.)

The dB values of DTI (Directional Threshold of Intelligibility: definition: Tønning 1971 *a*) were recorded without noise with the signal loudspeaker reproducing speech in four different positions in the horizontal plane (0° = in front, 90° = to the right, 180° = behind and 270° = on the left side of the person tested).

DTI was also determined during simultaneous stimulation with white noise: the signal loudspeaker was placed in each of the previ-

ously mentioned four positions in turn. For each of these positions, the noise loudspeaker was placed in the same four positions, resulting in a total of 16 different combinations of positions in noise.

Statistics in this article: Wilcoxon-Test for paired comparisons, level of significance 0.05 (References: Dixon & Massey 1957 Siegel, 1956). Electronic computer was used, the programs being taken from IBM's System 360 Scientific Subroutine Package (360A-CM-03X) Version III 1968.

The DTI values of each person were first determined without the use of hearing aid. Prior to the tests with hearing aid, the volume control of the aid was adjusted to an appropriate level by means of conversation with the operator under conditions being like for all the patients tested.

MATERIAL

Twenty right-handed experimental subjects (14 women and 6 men) from 26 to 73 years old (average age 56 years, median age 60 years) were examined. Pure tone audiograms were recorded for the frequencies 125 250 500 1000 2000 4000 6000 and 8000 Hz using a Madsen Electronics Audiometer Model OB60, calibrated according to British Standards.

The patients are listed in Table II. The hearing loss is given as PTA, i.e. Pure Tone Average: the mean of the hearing levels at the frequencies 500 1000 and 2000 Hz. The greatest difference between the PTA of the two sides in any one patient was 7 dB. Only 3 patients (nos. 3, 6 and 18) had used hearing aids before this investigation started.

Table I Table for transforming hearing loss in British Standard 1954 to ISO Standard 1964

Frequency Hz	125	250	500	1000	2000	4000	6000	8000
dB to be added to hearing loss in British Standard when transferring to ISO Standard	3.5	3.7	2.0	+0.9	0	0.0	7.9	5.5

Table II *Patients with bilateral hearing loss*

No.	PTA dxt.	PTA sin.	Type hearing loss	Sex	Age	Type hearing aid
1	45	43	Mixed	M	57	Aditone 620
2	40	35	Sn	♀	64	Orwidan 651E4
3	38	35	Sn	♀	63	Aditone 661
4	45	40	Sn	♀	61	Oricon 563U
5	32	28	Sn	♀	57	Oricon 830U
6	37	40	Sn	♀	99	Widex 641
7	41	38	Mixed	♀	46	Aditone 620
8	40	33	Mixed	♀	47	Oricon 560S
9	37	38	Sn	♀	26	Aditone 620
10	35	37	Sn	♀	73	Aditone 661
11	40	43	Sn	♂	65	Orwidan 651E12
12	45	38	Mixed	♀	44	Aditone 620
13	38	43	Mixed	♂	60	Aditone 620
14	37	40	Sn	♀	60	Aditone 620
15	37	40	Sn	♂	28	Aditone 620
16	42	40	Sn	♀	61	Aditone 675S
17	37	40	Sn	♀	60	Aditone 620
18	40	45	Sn	♂	60	Oricon 830S
19	43	42	Sn	♂	64	Widex 641
20	40	40	Sn	♂	64	Widex 641

PTA. Pure Tone Average: Mean of the hearing levels at 500, 1 000, and 2 000 Hz.

Sn: Sensorineural hearing loss.

Mixed: Combination of conductive and sensorineural hearing loss.

All patients were first examined without hearing aids. After an adaptation period of at least 2 months with a hearing aid, the DTI was measured with hearing aid. Patients 1-10 were first examined with binaural hearing aids after a minimum interval of a week they were examined with hearing aid on the right ear. Patients 11-20 were first examined with hearing aid on the right ear and after a minimum

interval of a week with binaural aids. For the 24 hours preceding the investigations the patient had only used hearing aid on the ear/ears due to be examined with aid.

RESULT

DTI without noise both without and with hearing aids

For each position of the signal loudspeaker the mean value for DTI (DTI) was calculated without hearing aid, with one hearing aid and with binaural hearing aids, and listed in Table III. It was found

1. Significantly better DTI with rightsided hearing aid compared with the DTI found without aid, for all the loudspeaker positions.

2. Significantly better DTI with binaural hearing aids compared with the DTI found without aid, for all the loudspeaker positions.

3. Significantly better DTI with binaural hearing aids than with monaural hearing aid for 3 of the 4 loudspeaker positions.

DTI in noise both without and with hearing aids

For each of the 16 combinations of signal and noise loudspeakers the arithmetic mean of the DTI values (DTI) without, with monaural, and with binaural hearing aids was calculated and listed in Table IV.

The test results are listed in Table V. They show

Table III *The influence of azimuth on the DTI without noise in 20 patients with bilateral hearing loss, recorded without with monaural and with binaural hearing aids*

DTI: The mean of DTI for each position of the signal loudspeaker

Position of signal loudspeaker	DTI without hearing aid	DTI with hearing aid dxt.	DTI with binaural hearing aids	Effect of hearing aid dxt. compared with no aid	Effect of binaural hearing aids compared with no aid	Effect of binaural hearing aids compared with hearing aid dxt.
In front	46.4	34.7	31.8	Improvement	Improvement	Improvement
Right ear	44.6	32.1	30.5	Improvement	Improvement	No effect
Behind	47.6	35.7	32.2	Improvement	Improvement	Improvement
Left ear	45.1	38.2	30.0	Improvement	Improvement	Improvement

Table IV The DTI values (=the arithmetic means of the DTI values) for the various combinations of signal and noise loudspeakers both without with monaural and with binaural hearing aids

Position of signal loudspeaker	DTI without hearing aid	DTI with hearing aid dist.	DTI with binaural hearing aids	DTI without hearing aid	DTI with hearing aid dist.	DTI with binaural hearing aids
Position of white noise loudspeaker- In front				Position of white noise loudspeaker- Behind		
In front	56.2	53.3	54.8	54.7	53.9	53.4
Right ear	53.7	52.5	50.4	52.2	52.0	50.8
Behind	56.4	54.6	54.5	56.3	55.8	54.8
Left ear	53.8	53.1	50.9	52.0	53.4	50.3
Position of white noise loudspeaker- Right ear				Position of white noise loudspeaker- Left ear		
In front	54.5	53.8	51.4	53.9	52.2	51.2
Right ear	56.6	56.3	53.4	51.0	48.3	47.7
Behind	55.3	55.0	51.5	55.0	51.9	51.1
Left ear	50.9	52.9	47.6	56.0	55.8	55.1

1 Significantly better DTI with right-sided hearing aid compared with DTI without aid for 2 positions. However the DTI was found to be worse with the apparatus for 2 positions.

2 Significantly better DTI with binaural hearing aids compared with DTI without hearing aid for 13 positions.

3 For 6 positions significantly better DTI was found with binaural hearing aids than with monaural hearing aid.

With a monaural hearing aid the listening position appears to be unsatisfactory when the signal source is facing the ear without a

hearing aid while simultaneously the source of noise is placed behind the head or aimed at the ear supplied with a hearing aid. Under these listening conditions significantly poorer DTI was found with monaural hearing aid than without an apparatus, whilst binaural hearing aids showed improvement of DTI.

Effect of hearing aids on individual DTI
For each patient the term DTI₀ indicates the DTI for all the 20 experimental positions without and with background noise. DTI₁₆ indicates the 16 positions with background noise.

Table V The effect of monaural and binaural hearing aids on the DTI for the 16 various combinations

Position of signal loudspeaker	Effect of hearing aid dist. compared with no aid				Effect of binaural hearing aids compared with no aid			
	Position of the white noise loudspeaker				Position of the white noise loudspeaker			
	In front	Right ear	Behind	Left ear	In front	Right ear	Behind	Left ear
In front	No	No	No	No	Improv	Improv	No	Impr
Right ear	No	No	No	No	Improv	Improv	Improv	Impr
Behind	No	No	No	No	Improv	Improv	Improv	Impr
Left ear	No	No	No	No	Improv	Improv	Improv	Impr

It was investigated

1 The effect on DTI₂₀ and DTI₁₀ of hearing aid on the right ear as compared with the values found without an apparatus.

2 The effect on DTI₂₀ and DTI₁₀ of binaural hearing aids as compared with the values found without an apparatus.

3 The effect on DTI₂₀ and DTI₁₀ of binaural hearing aids as compared with a right sided apparatus.

The results are shown in Fig. 1 and indicate that under our experimental conditions binaural hearing aids are more effective.

Use of the standard diagram

In evaluating the effect of the hearing aids, the standard diagram previously described (Tomning, 1971 *a* and *b* 1972 *a b* and *c*) may be used. In Fig. 2 this diagram is used to illustrate how hearing aids can influence the DTI values under various experimental listening conditions for patient 16.

1 Without hearing aid, the signal loudspeaker aimed at the left ear the noise loudspeaker aimed at the right ear

DTI without noise: 47 dB

DTI with noise: 49 dB.

Point plotted at A.

2. With hearing aid dxt., loudspeakers in the same positions.

DTI without noise: 33 dB.

DTI with noise: 49 dB

Point plotted at B.

3 With binaural hearing aids, the same

signal and noise loudspeaker

Test of binaural hearing aids compared with hearing aid dxt.

Position of the white noise loudspeaker

	Right ear	Behind	Left ear
1st	Improved	No	No
2nd	No	No	No
3rd	Improved	No	No
4th	Improved	Improved	No

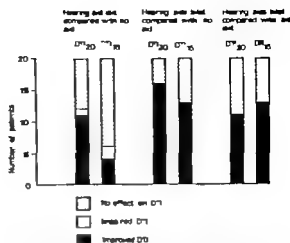


Fig. 1 The effect of monaural and binaural hearing aids on the 20 patients. DTI₂₀. The DTI both without and with background noise (4 loudspeaker positions without, and 16 with background noise). DTI₁₀. The DTI with background noise (16 loudspeaker positions).

positions of the loudspeakers

DTI without noise: 28 dB.

DTI with noise: 42 dB

Point plotted at C.

Point A lies farthest to the right in the diagram as an expression of the poorest DTI without noise. Point C lies farthest to the left as an expression of the best DTI without noise.

All the three points lie to the right of the area of the normal hearers, expressing that DTI found under our experimental conditions without noise is poorer than for the normal hearers.

Point C lies on the isoline farthest down in the coordinate system, indicating that, in this listening situation, the patient is less disturbed by noise than in the other two situations.

The points lie on isolines which cut the normal hearers area expressing that, in the actual listening conditions in noise, our patient has DTI values which do not differ from those of the 20 normal hearers. The isoline for point C has the lowest value and point C also lies farthest to the left in the coordinate

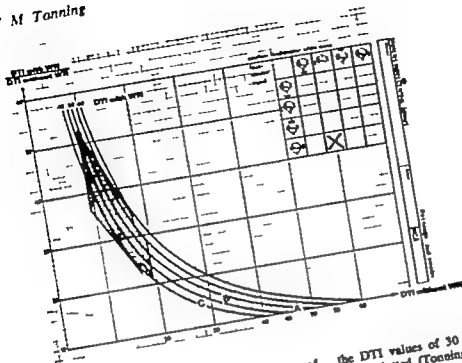


Fig. Coordinate system, illustrating the effect of monaural and binaural hearing aids under certain conditions. **Abscissa:** DTI without noise. **Ordinate:** DTI with noise divided by the corresponding DTI without noise i.e. with the signal loudspeaker in the same position. DTI in noise is also indicated. Curved lines. Shaded figure covers the area where 95% of

the DTI values of 30 normally hearing listeners are to be plotted (Tonning, 1971 a). DTI values for patient 16 are plotted in the diagram. A DTI without hearing aid. B. DTI with hearing aid on the right ear C. DTI with binaural hearing aids. For further explanation see text.

system as an expression of the best DTI values both with and without background noise impression of the influence both of the positions of the signals

An impression of the influence of the bearing aid and of the positions of the signal and noise sources on the comprehension of speech in the different conditions can be given by means of the diagram.

COMMENT

COMMENT

In no case have our calculations revealed significantly worse DTI values with binaural hearing aid treatment than with monaural. It must however be pointed out that our experimental conditions diverge considerably from listening situations in everyday life (Tonning 1971 b 1972 a b and c). The results therefore do not prove but suggest that binaural hearing aid treatment is advantageous.

For these measurable circumstances must be made the following constructive remarks by some

To these measurable circumstances must be added the purely subjective remarks by some

of the users of binaural hearing aids, the feeling of being able to hear with both ears. With binaural hearing aids they no longer have the impression of auditory imbalance. However the importance of such factors are hardly measurable in figures. In this connection it should be pointed out that Courtois (1972) reported good results of treating dizziness with hearing aids.

ZUSAMMENFASSUNG

ZUSAMMENFASSUNG

Zwanzig Patienten mit binokularen Hörschäden, resultierend von einem Reintone-Durchschnitt (PTA Pure Tone Average) von 28 dB Hörniveau. Die primäre Verständlichkeitsschwelle (DTI-Directional Threshold of Intelligibility) wurde mit und ohne Hörgerät (Grundgeräusch, mit und ohne Hörgeräteprozessor, Grundgeräusch, mit und ohne Hörgeräteprozessor) bestimmt. In 3 von den 4 Patienten ohne Hörgerät war das Hörniveau signifikant besser.

Ohne Hintergrundgeräusch, mit und ohne Hörapparat wurde die DII bei binazuralen Hörapparaten in 3 von den 4 Probanden des signalgebenden Lautsprechers signifikant bewertet.

gefunden als die DTI mit monauralen Hörapparaten. Mit Hintergrundgeräusch wurde die DTI mit binauralen Hörapparaten in 6 der 18 Kombinationen von signal- und geräuschgebenden Lautsprechern signifikant besser gefunden als mit monauralen Hörapparaten. Bei sämtlichen 20 experimentellen Hörverhältnissen fanden wir, dass die DTI mit binauralen Hörapparaten signifikant besser war als mit monauralen Hörapparaten bei 11 von 20 Patienten. Bei den 16 experimentellen Hörverhältnissen mit Hintergrundgeräusch fanden wir mit binauralen Hörapparaten eine signifikant bessere DTI als mit monauralen Hörapparaten bei 13 von 20 Patienten.

Die Resultate indizieren, dass binaurale Hörapparate vorzuziehen sind.

REFERENCES

- Belzile, M. & Marik, D. M. 1959 A clinical comparison of monaural and binaural hearing aids worn by patients with conductive or perceptive deafness. *Laryngoscope* 69 1317.
- Bender R. & Wigg, E. 1960. Binaural hearing aids for young children. *Voices Review* 62 113.
- Benzen, O. Grieson, O. & Jordaa, O. 1963 Bilateral hearing-aid treatment of 300 patients. *Acta Otol* 4 121.
- British Standard 1954 2497 *The normal threshold for pure tones by earphone listening* British Standards Institution, British Standards House, 2 Park St., London W1.
- Carhart, R. 1958 The usefulness of the binaural hearing aid. *J Speech Hearing Dis* 23 42 and *Trans Amer Acad Ophthal Otolaryng* Jan-Feb 1958, 120.
- Di Carlo, L. M. & Brown, W. J. 1960. The effectiveness of binaural hearing for adults with hearing impairments. *J Audiol Res* 1 35.
- Courtois, J. 1972. Deafness treated with hearing aids. *Scand Audiol* 1 13.
- Dirks, H. & Carhart, R. 1962. A survey of reactions from users of binaural and monaural hearing aids. *J Speech Hearing Dis* 27 311.
- Dixon, W. J. & Massey Jr F. J. 1957 *Introduction to statistical analysis*. McGraw-Hill, New York, Toronto, London.
- Haskins, H. L. & Hardy W. G. 1960. Clinical studies in stereophonic hearing. *Laryngoscope* 70 1427.
- Hedgecock, LaRoy D. & Sheris, B. V. 1958. A comparison of monaural and binaural hearing aids for listening to speech. *Arch Otolaryng* (Chic.) 68 624.
- Hirsh, I. J. 1950. The relation between localization and intelligibility. *J Acoust Soc Amer* 22 196.
- IBM 1968 *System/360 Scientific Subroutine Package (60A-CM-03X) Version III*.
- Jerger J. Carhart, R. & Dirks, H. 1961 Binaural hearing aids and speech intelligibility. *J Speech Hearing Res* 4 137.
- Jerger J. & Dirks, D. 1961 Binaural hearing aids. An enigma. *J Acoust Soc Amer* 33 537.
- Jordan, O. Grieson, O. & Benzen, O. 1967 Treatment with binaural hearing aids. A follow-up investigation of 1147 cases. *Arch Otolaryng* (Chic.) 85 105.
- Kodman, F. Jr. 1961 Successful binaural hearing aid users. *Arch Otolaryng* (Chic.) 74 88.
- Marik, D. M. & Aber W. 1958 A clinical evaluation of monaural and binaural hearing aids. *Arch Otolaryng* (Chic.) 67 606.
- Siegel, S. 1956 *Nonparametric statistics for the behavioral sciences*. McGraw-Hill, New York.
- Tønning, F. M. 1970. Directional audiometry I. Directional white-noise audiometry. *Acta Otolaryng* (Stockh.) 69 388.
- 1971a Directional audiometry II. The influence of azimuth on the perception of speech. *Acta Otolaryng* (Stockh.) 72 352.
- 1971b Directional audiometry III. The influence of azimuth on the perception of speech in patients with monaural hearing loss. *Acta Otolaryng* (Stockh.) 72 404.
- 1972a Directional audiometry IV. The influence of azimuth on the perception of speech in aided and unaided patients with monaural hearing loss. *Acta Otolaryng* (Stockh.) 73 44.
- 1972b Directional audiometry V. The influence of azimuth on the perception of speech in patients with monaural hearing loss treated with hearing aids (CROS). *Acta Otolaryng* (Stockh.) 74 37.
- 1972c Directional audiometry VI. Directional speech audiometry in patients with practical deafness in one ear and impaired hearing in the other ear treated with hearing aids. *Acta Otolaryng* (Stockh.) 74 206.
- Watson, N. A. 1942. Hearing aids: Uniform and selective: Monaural, diotic and binaural Air and bone conduction. *J Acoust Soc Amer* 13 335.
- Whitnall, E. 1964 Binaural hearing. *J Laryng* 78 1079.
- Whittle, L. S. & Delany M. E. 1966. Equivalent threshold sound-pressure levels for the TDH 39/MX41-AR earphone. *J Acoust Soc Amer* 39 1187.
- Wright, H. H. & Carhart, R. 1960. The efficiency of binaural listening among the hearing-impaired. *Arch Otolaryng* (Chic.) 72 109.

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THE EFFECT OF BRADYKININ ON EUSTACHIAN TUBE AND NASAL PATENCY

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Abstract. The patency of the Eustachian tube and nasal passages is decreased during inflammation due to vascular congestion. The vasodilation and increased capillary permeability due to histamine have been known for years and treated with antihistamines. Recent evidence of another endogenous substance involved in inflammation, bradykinin, ten times more active than histamine, has led to this study which showed the effectiveness of bradykinin in reducing the patency of the Eustachian tube and nasal passages. A comparison was made with an even more effective substance, eladolin, which is similar to bradykinin but does not occur in mammals. Two drugs, aspirin and Dilantin, that might have reduced inflammation by blocking bradykinin, were found to be ineffective. This may shed some light on the highly variable results obtained clinically when treating congested nasal passages and Eustachian tubes.

Bradykinin is similar to histamine in that it produces an inflammatory response but is more significant in that it is approximately ten times more potent and has no known blocking agent as antihistamines block histamine. This may have clinical import in otolaryngology where Eustachian tube and nasal congestion are common complaints.

There is good evidence that plasma kinins play an active role in inflammation by provoking (1) vasodilation (2) increased capillary permeability (3) accumulation and migration of leukocytes and (4) pain (Douglas, 1970). The effect like that of histamine is exerted on the small venules rather than on the true capillaries and involves separation of the junctions between endothelial cells (Majno et al., 1961).

Of the many vascular effects described for bradykinin, perhaps the most acceptable is that of venoconstriction, with selective constriction of A V shunts, increasing resistance to flow through the veins, resulting in a passive dilation of capillaries and arterioles (Selkja et al., 1971; Bobbin & Guth, 1968; DePasquale & Burch, 1966). The half life of bradykinin in blood is 0.27 minutes (McCarthy 1965). However bradykinin upon intra-arterial injection, reaches the venous receptors in sufficient quantities to induce a response. Moreover in the pathologic state as in tissue injury or inflammation, bradykinin may be released adjacent to or into postcapillary vasculature and cause venoconstriction (Bobbin & Guth, 1968). Blood contains both ingredients for kinin formation, kininogen and kallikrein. They are activated much as the circulating clotting factors, by various factors disturbing the equilibrium of the plasma. These include changes in pH or temperature, contact with glass and particulate matter (note Hageman factor in kinin formation scheme Fig. 1) collagen, microorganisms, allergic reactions and damaged tissues (Douglas, 1970; Eisen & Votaw, 1960).

Eladolin, an endopeptide first isolated from the salivary glands of two molluscan species, has the same effect as kinins, but is about ten times more active (Nakano & Asakura, 1966). It is similar to a "Substance P" in

This work was supported by NIH Research Grant
No. NS 90570-09 CMS

several times in each experiment to test the responsiveness of the animal. In most cases, nasal and Eustachian tube patency changes were measured in the same animal.

RESULT

The dose-response curve for bradykinin is compared with that of histamine in Fig. 3. A consistent feature of the drug response of the Eustachian tube is its seeming dependence on blood pressure change (Sheffield et al., 1970; Davis et al., 1970). This is also true in some cases for nasal patency (Fig. 4). Lower doses of bradykinin progressively decreased the tubal patency. Doses higher than about 0.005 $\mu\text{g/kg}$ resulted in reversal of the Eustachian tube patency responses. Thus, instead of leveling at a plateau at high doses, the responses often decrease in size as the dose increases producing a "peak" response. The dose at which the peak occurred was not consistent, nor was the threshold dose. Depending upon a particular animal's sensitivity these could shift \pm half a log unit. Most animals would develop rhinorrhea as doses increased. We were able to demonstrate the extreme potency of eledoisin, as one animal responded to an intra-arterial

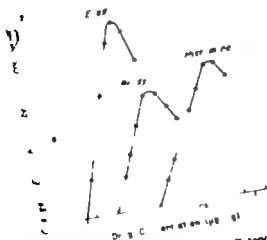


Fig. 3 Eustachian tube response. Patency decreased as Eustachian tube resistance increased. Drugs were injected into the common carotid. Data is averaged from seven dogs.

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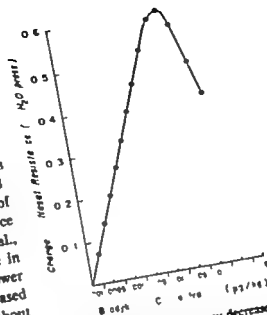


Fig. 4 Nasal response. Patency decreased as resistance to air flow increased. Bradykinin was injected into the common carotid. Data is averaged from three dogs.

dose of 0.25 picograms per kilogram of eledoisin.

Two drugs, aspirin and Dilantin, that might reasonably have reduced the inflammatory response by blocking bradykinin were found to be ineffective, both in doses comparable to the average dose in man (1 mg/kg) as well as ten times that dose.

DISCUSSION

The Eustachian tube and nasal patency changes induced by bradykinin are probably due to the effect on the blood vessels of the Eustachian tube and nasal mucosa. As bradykinin causes vascular congestion and mucosal swelling, patency decreases. The dose response curves that were obtained are in agreement with the relative potency of the drugs as found by other workers. That is, eledoisin is approximately ten times more potent than bradykinin which in turn is about ten times more potent than histamine.

It is of interest to note that a kinin-like substance has been isolated from the nasal

secretions of allergic patients who had been challenged with ragweed antigen (Dolovich et al, 1970).

ZUSAMMENFASSUNG

Im Entzündungsstadium wird die Durchlässigkeit der Eustachischen Röhre und der Nasenwege durch vaskuläre Kongestion beeinträchtigt. Es ist seit langem bekannt, dass Histamin Gefäßerweiterung und gesteigerte Permeabilität der Kapillaren hervorruft. Antihistamin-Präparate werden bei Behandlung benutzt. Neuerdings ist eine andere endogene Substanz, Bradykinin, die zehnmal wirksamer ist als Histamin, bekannt geworden. Die vorliegende Arbeit beschäftigt sich mit dem Einfluss von Bradykinin auf die Durchlässigkeit der Eustachischen Röhre und der Nasenwege. Bradykinin wurde zusammen mit einer ähnlichen, noch wirksameren Substanz, dem Eledoisin verglichen, das aber bei Säugtieren nicht vorkommt. Zwei weitere Medikamente (Aspirin und Dünatin), die u.U. entzündungshemmend wirken und deshalb die Wirkung von Bradykinin blockieren können, erwiesen sich als unwirksam. Diese Ergebnisse könnten zur Erklärung beitragen, warum man klinisch bei der Behandlung von verstopften Nasenwegen und der Eustachischen Röhre zu unterschiedlichen Ergebnissen kommt.

REFERENCES

- Bobbis, R. P. & Guth, P. S. 1968 Vasoconstrictive action of bradykinin. *J Pharmacol Exp Ther* 160 11.
- Davis, L. J., Sheffield, P. A. & Jackson, R. T. 1970. Drug-induced patency changes in the Eustachian tube. *Arch Otolaryng (Chic.)* 92 325.
- DePasquale, N. P. & Burch, G. E. 1966. Digital vasoconstrictor responses to intraarterial injections of bradykinin, kallikrein and eledoisin in man. *Circulation* 34 211.
- Dolovich, J., Beck, N. & Arbesman, C. E. 1970. Kinin-like activity in nasal secretions of allergic patients. *Int Arch Allergy* 38, 337.
- Douglas, W. W. 1970. Chap. 30 In *The pharmacology-*

- ical basis of therapeutics* (ed. L. Goodman and A. Gilman), Ed. 4 Macmillan, New York.
- Elsen, V. & Vogt, W. 1960. Plasma kininogenases and their activators. Chap. 3 in *Handbook of experimental pharmacology* (ed. E. G. Erdos), vol. 25 Springer Verlag, New York.
- Greenbaum, L. M., Fujii, Y. & Kim, K. S. 1966. Studies on the kinit-forming and inactivating enzymes in tissues and leucocytes. *Int Symp Vasoactive Polypeptides* Sao-Paulo, Brazil.
- Majno, G. et al. 1961. Studies on inflammation. II. Site of action of histamine and serotonin along vascular tree: topographic study. *J Biophys Biochem Cytol* 11 607.
- Marshall, E. A. 1967. Antagonism of anti-inflammatory drugs on bradykinin-induced increase of capillary permeability. *J Pharm Pharmacol* 19 617.
- McCarthy D. A., Potter D. E. & Nicolaidis, E. D. 1965. An in vivo estimation of the potencies and half-lives of synthetic bradykinin and kallikrein. *J Pharmacol Exp Ther* 148 117.
- Milner, N. J. 1955. The anatomy of the autonomic nervous system in the dog. *Amer J Anat* 96 285.
- Nakano, J. & Kozakari, T. 1966. Effect of bradykinin and eledoisin on the regional circulation. *Angiology* 17 333.
- Northover B. J. 1967. The effect of anti-inflammatory drugs on the deposition of colloidal carbon in the walls of venules. *J Path Bact* 94 204.
- Peiss, C. N. & Manning, J. W. 1964. Effects of sodium penicillinate on electrical and reflex activation of the cardiovascular system. *Circulation Res* 14 228.
- Pierce, J. V. 1968. Structural features of plasma kinins and kininogens. *Fed Proc* 46, 125.
- Sekiya et al. 1971. Studies on the vascular action of bradykinin. *Jap J Pharmacol* 21 87.
- Sheffield, P. A., Jackson, R. T. & Davis, L. J. 1970. Patency changes in the dog's Eustachian tube in response to alpha and beta adrenergic drugs. *Ann Otol* 79 117.
- Stovall, R. & Jackson, R. T. 1967. Prostaglandins and nasal blood flow. *Ann Otol* 76 1051.

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LETHAL INTRACRANIAL COMPLICATION FOLLOWING AIR INSUFFLATION WITH A PNEUMATIC OTOSCOPE

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CASE REPORT

Abstract Another case is reported of lethal intracranial complication following retrograde inflation through the external meatus. The cause of death was probably emphysematous detachment of the drum over the septum tympani and compression of the internal carotid artery. Insufflation of the middle ear should be carried out with extreme care in cases of secretory otitis media. Hyperbaric oxygenation could perhaps be of value in a case like this.

In recent years the treatment of secretory otitis media has included retrograde insufflation through the external auditory meatus to expel secretion from the middle ear and Eustachian tube using either a Politzer bag or a pneumatic otoscope (Siegle's). In 1963 Giske reported that he had used this method for several years without any complications, and in 1968 Courtois described over 1 500 carefully carried out insufflations without complication. A modern text-book (Scott Brown et al. 1965) describes paracentesis followed by insufflation of the tympanum through the external auditory meatus as an accepted form of treatment for secretory otitis media.

However as early as 1965 Ahren & Thulin published a case with "lethal intracranial complications following insufflation in the external auditory canal in treatment of serous otitis media and due to defects in the petrous bone". Fairman et al. published a similar case in 1968 with a lethal outcome due to air embolism after retrograde insufflation of the tympanum.

A 35-year-old previously healthy woman attended the out-patient clinic of the Ear, Nose and Throat Department, Södersjukhuset, Sweden on 10/3/69 because she has been troubled all winter by colds, and had reduced hearing in her right ear. A transudate was found in the right ear which was treated by paracentesis and nose drops. Nine days later the patient returned complaining of poor hearing as before. The tympanic membrane was found to be without perforations, yellowish and sluggish with a visible fluid level. Otherwise normal ear, nose and throat status. Paracentesis was carried out after surface anaesthesia with 2% tetracaine. A small quantity of yellowish fluid was tapped from the paracentesis opening and retrograde insufflation through the external auditory canal was carried out using Siegle's otoscope. The insufflation was not immediately successful but change to a better suited funnel resulted in passage of air. The impression obtained was, however, that the air had not passed through the Eustachian tube in the normal way so insufflation was discontinued after having introduced about one bag of air. The patient reported a bubbling sound in her right ear and a little pain in the right temporal region immediately afterwards. She lay down on a stretcher. Fifteen minutes after the insufflation she felt unwell and peculiar and

complained of severe pain in the right temporal region. After another 5 minutes she became confused and restless with commencing tonic convulsions, first on the right side later generalized. There was no bleeding from the ear.

An X-ray of the skull was taken as quickly as possible, but it was not possible to demonstrate intracranial air. The patient was intubated by an anaesthetist, and transported to The Neurosurgery Department, Karolinska Hospital in Stockholm. About an hour after the insufflation artificial respiration was necessary and the pulse became irregular. The systolic blood pressure varied between 175 and 145 mmHg. Angiography of the right internal carotid artery was carried out 3 hours after the insufflation. Markedly reduced passage of blood to the intracranial cavity was found. Only minimal amounts of contrast passed into very narrow intracranial vessels. An exploratory burr hole in the right temporal region revealed an extradural haematoma about 10 mm thick which was removed. Exploratory burr holes on the left side and in the posterior fossa showed no additional haematoma. The EEG was isoelectric and the patient was in a respirator. She died 8 days later.

Autopsy revealed relatively marked distention of the dura with no defects. The petrous part on the right side showed multiple punctiform reddish-brown changes very suggestive of bleeding. This was confirmed by microscopy. The brain was considerably oedematous and there was total encephalomalacia. No sign of intracerebral haemorrhage. There was no sign of infection. No sinus thrombosis. In the lungs there was extensive bronchopneumonia. There were no demonstrable bony defects in the petrous bone.

DISCUSSION

Ahrén & Thulin (1965) investigated 94 temporal bones and found that 21% had large or small defects of the tegmen tympani. In an additional 16% there was only a thin trans-

parent cortical bone covering the pneumatic cellulae of the tegmen tympani. In a model experiment with a Politzer bag a pressure of between 180 and 360 mmHg was recorded, which pressure was found sufficient to lift the dura off the tegmen tympani in one of two cases where there were defects in the tegmen.

Fairman et al. (1968) could not identify the pathway followed by the air from the tympanum to the carotid canal. They conclude that, as in the case of Ahrén & Thulin (1965) the air left the tympanum through one or more small hiatuses in the bony tegmen and then passed directly forwards beneath the dura to the roof of the carotid canal in the region of the foramen lacerum medium.

The cause of the symptoms in the present case was undoubtedly that air had propagated to the endocranium through pre-existent bone defects. The pathogenesis may be interpreted in different ways:

- 1 Separation of the dura from the petrous part thus severing blood vessels and resulting in formation of an extradural haematoma.

- 2 Separation of the dura and sudden compression of the cerebrum with diffuse brain damage as a consequence.

- 3 Air embolism as a result of air entering the venous system via ruptured thin walled vessels.

- 4 Compression of the internal carotid artery either by air penetrating into the bony canal of the artery or compression of the artery at the opening into the intracranial cavity by air passing between the petrosa and the dura.

The extradural haematoma shows that there has been loosening of the dura from the bone. An extradural haematoma of this size cannot however be the sole cause of the symptoms described. The volume of the bag used was 40 ml. The volume of air insufflated must therefore have been rather less than 40 ml. A sudden increase of intracranial pressure with severe compression of the brain cannot be excluded. However intracerebral bleedin

not demonstrated, as found by Chason et al. when studying the effect of a sudden increase in intracranial pressure in animal experiments. Air embolism was not considered likely as the blood pressure was approximately normal and there was no cyanosis. In the present case angiography showed markedly reduced passage of blood to the intracranial cavity. It is therefore reasonable to suppose that air has left the tympanic cavity through pre-existent bony defects in the tegmen tympani, and freed the dura from the petrosa, simultaneously severing small blood vessels and resulting in the extradural haematoma described. From there the air probably forced its way further under the dura to the region of the internal carotid artery compressing it.

The only treatment that might be of any value in a case like this is probably to give hyperbaric oxygenation within few minutes (Molvaer 1972). The air bubble in the tissue would decrease in volume as the pressure in the tissue increased, according to the law of Boyle-Mariotte. The volume of the air bubble would shrink also because the amount of physically dissolved nitrogen increases with the pressure. By inhalation of pure oxygen the diffusion of nitrogen from the bubble would increase. At a pressure of 3 atm there would be 6.6 ml O₂ per 100 ml artery blood which is sufficient oxygen supply for the resting organism.

Such treatment must be started before irreparable brain damage has occurred. However the possibility for such treatment is not at hand in many places.

COMMENT

In 1948 Hagness published a case with a lethal outcome as a result of air embolism after Eustachian catheterization and insufflation and he refers to four other cases in which the same treatment also resulted in death

(Wanibucki, 1928 Savage 1839- Blegvad, 1939) Hagness refers to Behrend who in 1920 had a case with lethal outcome after catheterization with insufflation on a 3 years old patient. He also mentions a number of cases in which there was loss of consciousness and transient cessation of pulse and respiration (Savage, 1839 Blegvad, 1939- Ramberg, 1940). Thoruvald deals with this complication in his text-book.

Ahrén & Thulin (1965) also describe a case of air embolism resulting in death after catheterization and insufflation. Both retrograde insufflation and Eustachian catheterization with inflation may obviously be dangerous procedures and should be carried out with extreme care, if indeed these methods of treatment should be used at all in patients with secretory otitis media.

ZUSAMMENFASSUNG

Darstellung des klinischen Verlaufes bei einem Fall mit letalem Ausgang nach retrograder Luftinfusion in den Ohrtrichter im äußeren Gehörgang. Die Todesursache war wahrscheinlich eine embolische Durchblutung von Tegmen tympani und Kompression der Arteria carotis interna. Luftdurchblutung des Mittelohres darf bloß mit größter Vorsicht angeführt werden. Überdruckbehandlung mit Sauerstoff wäre vielleicht wertvoll in diesem Falle.

REFERENCES

- Fairman, H. D. Brown, N. J. & Hallpike, C. S. 1963. Air embolism as a complication of inflation of the tympanum through the external auditory meatus. *Acta Otolaryng* (Stockh.) 66 65.
- Hagness, I. 1948. Letal infektion etter tubage. *Arel Med* 40 1781.
- Molvaer O. I. 1972. Personal communication.
- Scott-Brown, W. G. Ballantyne, J. & Groves, J. 1965. *Diseases of the ear nose and throat* 2nd ed., vol. 2, p. 437. Butterworths, London.
- Ahrén, C. & Thulin, C. A. 1965. Letal intracranial complications following inflation. *Acta Otolaryng* (Stockh.) 60 407.
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AUDIOLOGICAL ASPECTS OF ACUTE OTITIS MEDIA

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(Received April 17 1972)

Abstract. An unselected material of 136 patients with signs and symptoms of acute otitis media were audiometrically tested with both air and bone conduction. The audiograms were made at intervals during the healing period. The first audiogram was taken before any kind of treatment. No sensorineural hearing impairments were registered. The air-bone gap averaged 20 dB initially but was closed in most cases in 20 days. No permanent hearing impairments were noted. No major differences were noted between the audiograms of patients with spontaneous rupture, myringotomy and no myringotomy.

Acute otitis media is a frequent complication of upper respiratory tract infections, especially in children. It is nowadays seldom combined with serious complications as it used to be before the introduction of chemotherapeutics and antibiotics. Transient hearing impairment is, however still reported after an attack of acute otitis (Olmsted et al. 1964 Neil et al. 1966) although it is seldom permanent (Stickler et al., 1967). There are unfortunately few studies with audiometric measurements in the acute stage of the infection before any treatment and with a consequent follow-up during the restitution period.

Acute otitis media must not be considered healed until hearing is normal. Therefore, it was thought of interest to include audiological measurements while studying etiological, clinical and serological aspects on acute otitis media.

MATERIAL AND METHOD

The investigation was performed during the period September-December 1968 and comprised 237 out-patients with symptoms and signs of acute otitis media in the Otolaryngological Department, Sahlgren's Hospital, Göteborg.

The age distribution is stated in Fig. 1. 136 patients over 3.5 years of age were audiometrically tested with both air and bone conduction. Air and bone conduction audiometry was performed with a Campex DA II audiometer calibrated according to ISO standard, 1964. The tests were made in a sound-proof room. After the otological examination bacterial and blood samples were taken for analysis (Herberts et al., 1971).

All patients were treated with either of two penicillin preparations.¹ Nose decongestants were also prescribed and the therapy was continued for 10 days. No therapeutical differences were noted between the two preparations and therefore the material was analysed as a single entity (Herberts et al., 1971).

The first audiogram was made immediately after the otitis had been diagnosed and before the patient had received any kind of treatment. Thus, the myringotomies, if any were made after the test. The second audiogram was performed after 9-11 days of penicillin treatment. The third audiogram was made after another 10 days on patients considered not healed at the first control. Some of these had continued antibiotic treatment for more than 100 days. In those cases, the drug was chosen in accordance with the sensitivity test of the organism isolated at the first visit.

The purpose of this investigation was to

¹ Penicillin-V and alpha-azido-benzyl-penicillin (azido-cillin) which latter has the same antibacterial spectrum as penicillin-G but a higher effect in vitro against *H. influenzae*. The preparations have kindly been placed at our disposal by Astra Läkemedel AB, Södertälje, Sweden (Forsgren, U 1968).

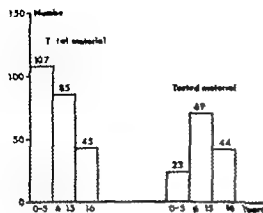


Fig 1 Age distribution.

study the hearing function in cases of uncomplicated otitis media. In order to elucidate the importance of spontaneous rupture and myringotomy respectively on hearing, the patients were divided in the following three groups:

- 1 Patients with spontaneous rupture (42).
- 2 Patients in whom myringotomy was performed (50)
- 3 Patients in whom myringotomy was not indicated (44)

RESULT

The mean hearing levels of the frequencies 250-8000 Hz were recorded on 136 patients (Fig. 2). It can be seen that the results of the bone conduction tests (8000 Hz not tested) did

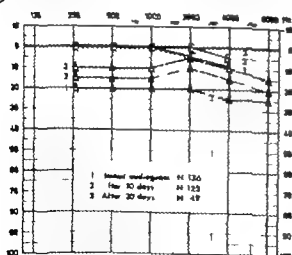


Fig 2 Mean audiogram of the affected ear for the total patient material.

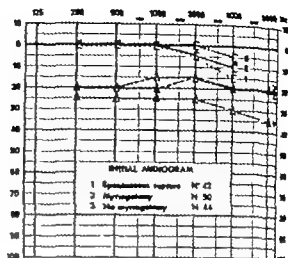


Fig 3 Mean initial audiograms for the affected ear in patients with spontaneous rupture, myringotomy and no myringotomy.

not vary at all under the infection period. The air-bone gap at the first test averaged 20 dB for all frequencies up to 4000 Hz (Fig. 3). After 10 days of penicillin therapy the gap was considerably reduced (Fig. 4). Forty-nine patients were considered not healed at this control and were again tested after another 10 days (Fig. 3). At this occasion the air-bone gap was closed and the hearing considered to be normal in all but 6 cases with secretory otitis media.

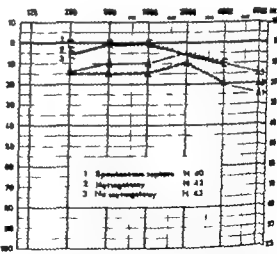


Fig 4 Mean audiograms after 10 days for the affected ear in patients with spontaneous rupture, myringotomy and no myringotomy.

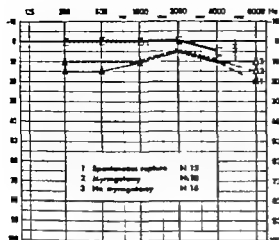


Fig. 5 Mean audiograms after 20 days for the affected ear in patients with spontaneous ruptures, myringotomy and no myringotomy

The mean audiogram at different times of the three groups of patients with spontaneous ruptures, myringotomies and no myringotomy are presented in Figs. 3-5. The average bone conduction curves for the different groups were similar and without changes during the infection. There are astonishingly small differences in the hearing thresholds between the patients with spontaneous rupture, myringotomy and those who were only antibiotically treated. The only notable point is that the pa-

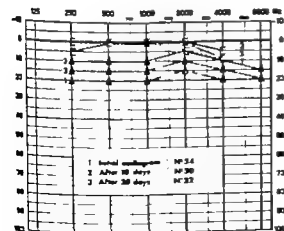


Fig. 6 Mean audiograms for the affected ear in patients with isolation of *Haemophilus influenzae*

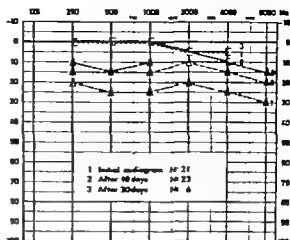


Fig. 7 Mean audiograms for the affected ear in patients with isolation of *Diplococcus pneumoniae*

tients with spontaneous rupture had a somewhat lower initial air conduction curve than the other two groups.

Neither could any differences be found between the audiograms of patients with different bacterial isolations (Figs. 6-7).

Six patients as mentioned before had a residual secretory otitis media with large amounts of highly viscous transudate. The mean audiograms of these patients (Fig. 8) showed a more pronounced air-bone gap at all frequencies, even at the first control, compared with the

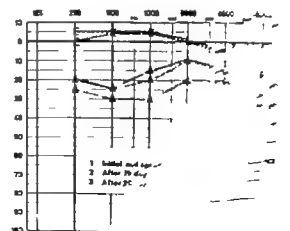


Fig. 8 Mean audiograms for patients with residual secretory otitis media

total material. The air-bone gap of these subjects was reduced slowly. The hearing was normalized first after treatment with antihistamine and after myringotomy.

DISCUSSION

The air conduction thresholds of patients with acute otitis media were only moderately elevated in the acute stage. There were surprisingly small differences between the thresholds of those with bulging ear-drums (later myringotomized) and of those who were treated with only antibiotics. The initial air-bone gap was about 20-25 dB for all frequencies. The bone conduction thresholds were all within normal limits and no sensorineural impairments appeared. After 10 days most of the lapse of a normal hearing independent of the lapse of the otitis. The moderate to slight impairment and the rapid normalization of the hearing may be due to the short lapse of the otitis before therapy and the quick restitution of the mucous membrane of the middle ear, mastoid and the Eustachian tube under adequate antibiotic treatment.

In an earlier paper on acute otitis media the patients were divided into three groups according to the therapeutic results (Herberts et al. 1971).

The audiograms of the therapeutic groups were compared. No major differences could be found between the patients with an uneventful healing and those regarded as therapeutic failures.

ZUSAMMENFASSUNG

Ein nicht ausgewähltes Material von 136 Patienten mit Zeichen und Symptomen einer akuten Otitis-Media sind audiologisch, bezüglich ihrer Luft- und Knochenleitung untersucht worden. Diese audiologischen Untersuchungen sind während der Heilungsperiode in verschiedenen Zeitabständen wiederholt worden. Die erste audiologische Untersuchung wurde vor Beginn irgendeiner Behandlung vorgenommen. Es konnte keine Verschlechterung der Gehörschwellenempfindlichkeit gemessen werden. Der Abstand der Meßwerte zwischen Luft- und Knochenleitung war zu Anfang 20 dB derselbe schloß sich jedoch in den meisten Fällen im Laufe von 20 Tagen. Es konnte außerdem keine fortbestehende Verschlechterung des Gehörs registriert werden. Zwischen den Audiogrammen der Patienten, mit Spontaner Ruptur Myringotomie und nicht Myringotomie konnten keine großen Unterschiede festgestellt werden.

REFERENCES

- Forsgren, U. 1968. Further in vitro studies of nido-cillin against haemophilus. In *Antibacterial agents and chemotherapy* p. 449.
- Herberts, G., Jeppsson, P.-H., Nylen, O. & Brander-Helander, P. 1971. Acute otitis media. Etiological and therapeutic aspects on acute otitis media. *Pract Otorhinolaryng (Basel)* 33: 191.
- Neil, J. F., Harrison, S. H., Morbey, R. D., Robinson, G. A., Tate, G. M. T. & Tate, H. T. 1966. Deafness in acute otitis media. *Brit Med J* 1: 75.
- Olmsted, R. W., Alvarez, M. C., Moroney, J. D. & Evernden, M. 1964. The pattern of hearing following acute otitis media. *J Pediatr* 65: 252.
- Stöckler, G. B., Rubenstein, M. M., McBeas, J. B., Hedgecock, L. D., Hugstad, J. A. & Grütting, T. 1967. Treatment of acute otitis media in children. IV. A fourth clinical trial. *Amer J Dis Child* 114: 123.
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MEDIASTINOSCOPIC OBSERVATIONS OF METASTATIC SPREAD IN PULMONARY CARCINOMA

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(Received June 28, 1972)

Abstract Mediastinoscopy was made in 338 patients with lung cancer and biopsies were taken from paratracheal, parabranchial and carinal lymph nodes. Positive biopsies were obtained in 159 cases (47%). In 168 right lung tumors, metastases were found ipsilaterally in 49% and contralaterally in 20%. Of 170 left lung tumors, 45% showed metastases ipsilaterally and 25% also on the contralateral side. Contralateral metastases from the right upper lobe were rare (7%). Histologically epidermoid carcinoma showed the lowest number of metastases (34%) while the microcellular anaplastic type had metastases in 81%. When the mediastinal lymph nodes become involved, curative surgery is very rarely useful and radio- or chemotherapy should be considered.

Rouvière's (1932) findings concerning the normal pulmonary lymph flow indicated that the flow from the right lung is directed straight to the tracheo-bronchial lymph nodes or to carinal nodes and from there to the paratracheal nodes. From the left side the lymphatic channels pass either to the tracheo-bronchial and anterior mediastinal lymph nodes, or to carinal and from there to the right paratracheal nodes. Consequently in malignant disease, metastases from the left side have been considered to occur frequently to the right whereas metastases to the left paratracheal nodes from the right lung are believed to be rare.

In his extensive studies, Nohl (1962) and Nohl-Oser (1971) presented detailed data on metastatic nodes found in the resected specimens and at mediastinoscopy. Nohl-Oser emphasized Borrie's concept (1952) that the lymphatic stump distal to the upper lobe bronchi

is the main site of early metastases, from which area further spread occurs to the tracheo-bronchial and carinal nodes. In his mediastinoscopic examination of 316 patients with right-sided lung cancer metastases to the left occurred in 3.8% whereas there was spread to the ipsilateral side in nearly 50%. In a total of 248 cases with carcinoma of the left lung, 24% showed metastases to the regional tracheo-bronchial nodes and 15% to the contralateral paratracheal nodes. Rouvière's data were thus confirmed in general. On the other hand, Greschuchina & Maassen (1971) reported a clearly higher incidence of metastatic spread from right to left, and in their series of 479 cases of carcinoma of the right lung, 11% showed contralateral metastases. This spread most frequently (26%) occurred from the middle lobe and least frequently (5%) from the lower lobe. Contralateral tumors were found in 24% of 427 cases with carcinoma in the left lung. They somewhat less frequently arose from the upper lobe than from lower lobe tumors (33%).

Our earlier studies (Palva, 1964; Palva et al., 1968) of metastatic spread based on mediastinal biopsies were more in line with the data of Greschuchina & Maassen (1971). In this paper we will evaluate our findings in the present material of 338 cases of pulmonary carcinoma subjected to mediastinoscopy and lymph node biopsy from various sites during the period 1964-1971.

Table I Localization of primary tumor

Histologic classification	Right lung					Left lung					Total	Total
	Main bronchus	Upper lobe	Middle lobe	Lower lobe	Total	Main bronchus	Upper lobe	Lingula	Lower lobe	Total		
Anaplastic carcinoma		10	7	4	21	4	15	3	8	30	51	117
Microcellular		35	11	10	57	6	29	1	24	60	119	25
Other	1	27	13	9	50	10	30	6	14	60	119	5
Epidermoid carcinoma	10	10		3	23	2	6	1	2	11	21	5
Adenocarcinoma		1		7	8				1	1	2	21
Alveolar cell carcinoma		6			6				2	2	8	170
Non-defined			31	37	68	22	86	11	51	170	338	
Total	11	89										

Figures referring to middle lobe include tumors of the stem bronchus.

MATERIAL AND RESULTS

Table I shows the localization of the primary tumors of the right and left lung compared with the histological type. The cases were classified into four groups by the site affected in each lung (main bronchus, three principal lobes) the lingular segments being listed separate from the upper lobe on the left side. Tumors arising in the right stem bronchus distal to the upper lobe bronchi down to the level of the upper rim of the superior lower lobe bronchus were included among the right middle lobe lesions.

Table II gives the number of positive and negative biopsies and the main method of treatment. The number of positive biopsies, 159 represented 47% of the total material. The majority of these patients (147) were treated conservatively. In 12 cases, operative treatment was attempted, but in none was curative resection possible. In the 179 cases with negative mediastinoscopy operative treatment was the method mainly used (122 cases or 68%). In the remaining cases conservative treatment was adopted for a variety of reasons. Among the cases with negative biopsies during mediastinoscopy there were 25 (14%)

Table II Findings at mediastinoscopy related to treatment

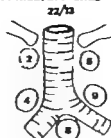
	Tumor type		Epidermoid	Adenoc.	Alveolar	Non-defined	Total
	Anaplastic	Microc.					
Positive biopsy	39	63	39	13	1	4	159
Lobectomy	1	2	2	2			6
Pneumonectomy			2	1			3
Thoracotomy		1	2	1		4	147
Conservative treatment	38	60	34	10	1	17	179
Negative biopsy	12	54	80	12	2		59
Lobectomy	3	23	24	7	1		43
Pneumonectomy	5	12	25	2		5	20
Thoracotomy	2	5	6				57
Conservative treatment	2	14	25	3	1	12	338
Total	51	117	119	25	5	21	

which carcinomatous lymph nodes were found at resection. However the majority of these were situated either in the lymphatic stump, or posterior or anterior to the mediastinoscopy level, or at the periphery. In 5 cases (3%) metastases were demonstrated in the area clearly accessible to the mediastinoscope.

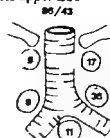
Figs. 1 and 2 show metastatic spread from the areas indicated in Table I. From the right upper lobe, metastases to the right tracheo-bronchial area were the rule, 47 (53%) of 89 cases, but to the left they were very rare, totalling only 2 cases (2%). For metastases from the right main bronchus these figures were respectively 55% and 18% from the middle lobe 48% and 26% and from lower lobe carcinoma 41% and 14%. Metastasis to the contralateral side was thus considerably more frequent from the three other sites than from the upper lobe.

For the left lung, metastases to the right side arose more frequently from the main bronchus (5 cases or 23%) than from the up-

Left main bronchus



Left upper lobe



Lingula



Left lower lobe



Superior mediastinal spread

Fig. 2 Number and sites of malignant nodes discovered on mediastinoscopy in tumors of the left lung.

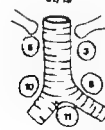
Right main bronchus



Right upper lobe



Right middle lobe



Right lower lobe



Superior mediastinal spread

Fig. 1 Number and sites of malignant nodes discovered on mediastinoscopy in the right lung. Figures indicate the total number of cases and the number with metastatic nodes for each of the four parts.

per (9 cases or 10%) or lower (5 cases or 10%) lobe. Ipsilateral metastases from the left upper lobe were just as common as on the right side (49%). Metastases from the lingular segments were found in this series on the ipsilateral side only (17%) but the series is small.

Figs. 3 and 4 relate metastases to histological type of the tumor. Metastatic spread from the right side was least frequent in the case of epidermoid carcinoma (34%) adenocarcinoma metastasized in 50% microcellular in 81% whereas involvement of nodes by other anaplastic carcinomata occurred in 61%. Contralateral metastases were comparatively rare in cases of adenocarcinoma and epidermoid cancer (7% and 5% respectively) while the microcellular type showed contralateral spread in 19% and other anaplastic types in 14%.

Metastases from the left lung, from epidermoid carcinoma were also less frequent (32%) than from the other three types (microcellular 73% other anaplastic 47% adenocarcinoma 55%). Contralateral metastases occurred in all tumor types but here too epider-

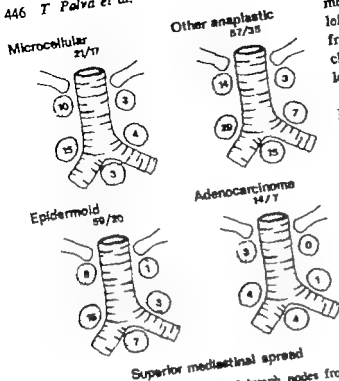


Fig. 3 Distribution of infiltrated lymph nodes from tumors of the right lung in relation to histologic type of tumor

moid carcinoma showed the lowest figure (6%) the other types varying from 12% to 16%.

Metastatic spread, demonstrated as positive mediastinal biopsies for the right and left lungs, is shown in Fig. 5. Of the 168 right lung tumors, metastases were found in 83 cases (49%), and of these 17 (20%) showed metastases on the contralateral side. Of the 170 tumors of the left lung 76 (45%) had metastases and 19 (25%) of these had metastases to the contralateral right side.

COMMENT

The results of this series of 338 cases of lung cancer are in agreement with our earlier data and confirm the findings of Greschuchna & Maassen (1971) indicating considerable contralateral spread also from the right lung. In our cases this was slight from the right upper lobe lesions (2%) spread from the middle lobe and stem bronchus showing the highest figure (26%) and from the lower lobe an inter-

mediate figure (14%). Metastases from the left lung towards the right occurred most often from the main bronchus tumors (23%) and clearly less often from the upper and lower lobes (10%).

Even admitting the general correctness of Rouvière's concept, it is a fact that metastatic spread from right to left occurs in a fair number of those cases in which the route from the lymphatic sump leads to the carinal region. This must be taken into account particularly when patients with cancer in the middle lower lobes are being evaluated for operative procedures.

It is clearly established by now that, if metastases have extended beyond the lymphatic sump representing stage 3 in the Ss Nohi classification there is a general lymphatic system involvement. The disseminated lymphatic system involvement pointed out by Nohi-Oser is not limited to lymph nodes but involves the lymphatic vessels between them. This means that because of nodal involvement all lymphatic c-

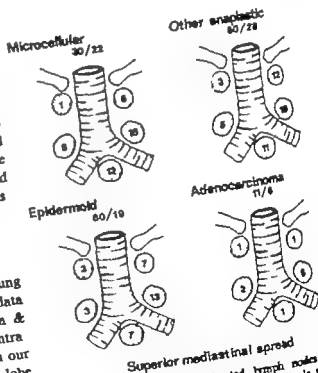
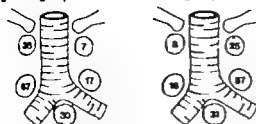


Fig. 4 Distribution of infiltrated lymph nodes in tumors of the left lung as related to histologic type of tumor

Right Lung 108/83 cases Left Lung 170/76 cases



Superior mediastinal spread

Fig 3 Total number and sites of invaded lymph nodes found by mediastinoscopy in right and left lung carcinoma.

along the spread route must be infiltrated. It implies further as stated for instance by Nohl-Oser (1971), Palva (1964), Palva et al. (1968) Paulson & Urschel (1971) and by ourselves (1971) that resection, even though it might be technically possible is extremely seldom of benefit. Equally good or better results are achieved by modern radio- or chemotherapy without postoperative mortality and prolonged morbidity. If resection is being planned, Kirschner's (1971) suggestion to give first a full dose of radiotherapy for eradication of the mediastinal disease, combined later with resection of the primary tumor may be valuable but has yet to be proved by a large enough series of cases.

ZUSAMMENFASSUNG

Mediastinoskopie wurde bei 338 Patienten mit Lungenkarzinom durchgeführt und Biopsien wurden aus den paratrachealen, parabronchialen und Bifurkationslymphknoten entnommen. Histologisch positive Befunde begegnete man in 159 Fällen (47%). Bei 168 Tumoren der rechten Lunge wurden ipsilaterale Metastasen in 49% und kontralaterale in 20% der Fälle gefunden. Für 170 Karzinome der linken Lunge lag

die ipsilaterale Metastasenrate bei 45% und die kontralaterale bei 25%. Die kontralateralen Metastasen vom rechten Oberlappen waren selten (2%). Von den histologischen Typen zeigte das Plattenepitheliarkarzinom die niedrigste Metastasenhäufigkeit (34%) und das kleinzellige Bronchuskarzinom die höchste (81%). Bei Tumoren, die mediastinale Metastasierung zeigen, ist chirurgische Therapie sehr selten erfolgreich. Diese Fälle müßte man durch Bestrahlung oder Chemotherapie behandeln.

REFERENCES

- Borrie, J. 1952. Primary carcinoma of the bronchus: prognosis following surgical resection. *Ann Roy Coll Surg Engl* 10 165.
- Greschuchna, D. & Massen, W. 1971. New observations of lymphatic spread of bronchogenic carcinoma. In *Mediastinoscopy* (ed. O. Jepsen & H. R. Sørensen). Odense University Press, Odense.
- Kirschner P. 1971. Mediastinoscopy as a guide to pre-operative radiotherapy and surgery in lung cancer. In *Mediastinoscopy* (ed. O. Jepsen & H. R. Sørensen). Odense University Press, Odense.
- Nohl, H. C. 1962. *The spread of carcinoma of the bronchus*. Lloyd Lulu, London.
- Nohl-Oser H. C. 1971. The lymphatic spread of carcinoma of the bronchus. In *Mediastinoscopy* (ed. O. Jepsen & H. R. Sørensen). Odense University Press, Odense.
- Palva, T. 1964. *Mediastinoscopy*. S. Karger AG Basel.
- Palva, T. Palva, A. & Kkry, J. 1968. La médiastinoscopie. *Ann Otol* 85 531.
- Paulson, B. L. & Urschel, H. C. 1971. Selectivity in the surgical treatment of bronchogenic carcinoma. *J Thor Cardio Surg* 62 554.
- Rouvière, H. 1932. *Anatomie des lymphatiques de l'homme*. Masson et Cie, Paris.
- Salzer G. 1951. Vorschlag einer Einteilung des Bronchien-Karzinoms nach pathologisch-anatomisch-klinischen Gesichtspunkten. *Wien Med Woch* 101 102.

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THE NORMAL ELECTROMYOGRAM IN HUMAN CRICOTHYROID MUSCLE

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(Received July 1 1972)

Abstract The electromyographic pattern in 17 cricothyroid muscles of 13 healthy subjects has been determined. The EMG recordings were made with a concentric needle electrode, recording area 0.03 mm². In the majority of the muscles there was continuous activity during quiet breathing throughout the phases of respiration. Some motor units fired with a steady frequency whereas in the others the discharge frequency varied with the respiratory phases, some showing an increase during inspiration, others during expiration. Some 20 motor unit potentials were identified from each muscle and, in all, 338 motor unit potentials were analysed. Most of the action potentials were di- or triphasic. Polyphasic potentials occurred in 1% of the total number of potentials. The mean duration of the action potentials was 3.99 ± 1.40 ms. The incidence of potentials < 2 ms in the whole material was 5% and varied in individual muscles between 0 and 16%. The mean amplitude was $394 \mu V$ and the 10th and 90th percentiles were $150 \mu V$ and $800 \mu V$ respectively. The maximal amplitude of the interference pattern during deglutition was usually in the range of 1 200-2 000 μV .

As shown in previous studies of the electromyographic pattern in vocal cord paresis (Haglund et al., 1970 1972) neurogenic lesions may be established by several different criteria, viz. an increased incidence of potentials of short or long duration, a high frequency of potentials with high amplitude or an altered incidence of polyphasic potentials, as well as changes in the interference pattern during phonation or deglutition. Thus, in order to establish electromyographic criteria that may facilitate diagnosis in cases of disturbance of motor functions of the

larynx, a rather extensive knowledge of the variability in duration, amplitude and shape of the motor unit potentials of healthy muscles is needed. Such data from recordings in the vocal muscles are available (Kinnison et al., 1969) and have been the basis for electromyographic investigations in cases of traumatic paresis of the recurrent nerve (Haglund et al., 1970) and of idiopathic vocal cord paresis (Haglund et al., 1972).

The motor control of the laryngeal muscles is exercised by the recurrent and the superior laryngeal nerve, the latter innervating only the cricothyroid muscle. To render possible a more complete evaluation of motor disturbances in the larynx, a determination of the electromyographic pattern in the cricothyroid muscles should therefore preferably be added to the studies of the electromyographic activity of the vocal muscles. A conclusive evaluation of possible lesions in the cricothyroid muscles of patients with voice disturbances is, however, hampered by the lack of sufficient data to allow an estimation of the significance of electromyographic findings, since only the mean duration of samples of few potentials from individual muscles are known (Faaborg-Andersen 1957). The present study of shape, amplitude and duration of a significant number of motor unit potentials, from each of a series of healthy muscles was therefore undertaken. The resulting data provide a basis for the use of electromyography in the diagnosis of neuro-

This work was supported by the Swedish Cancer Society (Project No. 502-B71-01P).

genic lesions in the superior laryngeal nerve. As will be shown in a separate paper they facilitate evaluations of the pathogenesis of voice disturbances of unknown origin.

MATERIAL AND METHOD

Seventeen muscles from 13 healthy individuals were examined. The group consisted of 5 men and 8 women of mean age 31 years (range 23-63 years). None of the subjects had any previous history of voice disturbance. Indirect laryngoscopy was performed in all subjects and revealed a normal position and normal motility of the vocal cords.

Electromyographic recordings were made with a concentric needle electrode (Diss type 9013 K 32, needle length 43 mm, outer diameter 0.65 mm, leading-off surface of the platinum core 0.03 mm² and bevel angle 15°). The recordings were made between the centre core and the outer needle. A chest electrode placed between heart and larynx connected the subject to earth. The recordings were fed to a differential amplifier with an input impedance of 3 megohms and an essentially linear frequency response between 4 Hz and 10 kHz. The output was connected to an audiomonitor and a loudspeaker as well as to an oscilloscope. The potentials were photographed on moving film, from which all measurements were made. Analyses of shapes, durations and amplitudes of the action potentials were made only when at least 2 identical recordings from the same motor unit were obtained and the onset and end of the potentials could be well defined.

In order not to interfere with the action potentials to be recorded, no kind of anesthetic was given to the subjects. Nor did they receive premedication. The skin was pierced in the midline over the cricoid arch at the level of its superior margin. The tip of the needle was inserted until it made contact with the perichondrium. The needle was then directed laterally and slightly upwards and was allowed to follow over the cranial margin of the cricoid arch laterally. It was sometimes possible to

feel the medial margin of the cricothyroid muscle with the palpating hand, and often also to feel when the needle penetrated the muscle. This mode of inserting the electrode enables an exploration to be made of a large portion of the muscle. Unintentional recordings from the sternothyroid and sternohyoid muscles were avoided by bringing the needle into immediate contact with the perichondrium of the cricoid arch. When the needle penetrates the cricothyroid muscle, the oscilloscope beam displays a muscle activity pattern. Motor units were identified by carefully advancing and rotating the needle. The needle was never advanced further than 10 mm into the muscle, but sometimes several insertions were made. By this method it was possible as a rule to identify at least 20 motor unit potentials from each muscle investigated. The subjects did not feel any pain when the muscles were explored, but experienced a certain discomfort when the needle touched the perichondrium. None of the subjects have noticed any degree of hoarseness since the examination. On the day following the exploration some of them felt a transient tenderness in the investigated muscles, which was most prominent during swallowing. Otherwise no discomfort was experienced.

RESULT

Activity pattern

The activity in the muscle was studied both during quiet respiration and during activation. During quiet breathing there was continuous activity in the cricothyroid muscle in 12 of the subjects investigated in spite of every effort to achieve relaxation (cf Weddell et al., 1944; Faaborg-Andersen, 1957). Some motor units fired at a steady frequency whereas the frequency of others varied with the respiration. Some of the latter motor units were active only during one of the respiratory phases, while others fired continuously modulating their frequency with the respiratory rhythm. Most units fired with a frequency of 10-20 per sec. The highest rate observed was 33 per sec. In

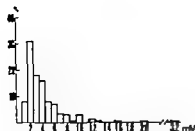


Fig. 2 Histogram showing the distribution of the amplitudes of 338 action potentials recorded from 17 normal cricothyroid muscles.

tudes of all potentials have been plotted. As will be seen the distribution is skewed. On account of this it has been considered more reliable to describe the 10th and 90th percentiles as a basis for later evaluations of pathological conditions. The 10th percentile has been calculated to be $150 \mu\text{V}$ and the 90th to be $800 \mu\text{V}$ and it follows that 80% of all potentials had amplitudes within this range. In 15 of the 17 muscles tested more than 80% and in the remaining 2 muscles between 70% and 80% of the potentials had amplitudes within the range $150\text{--}800 \mu\text{V}$. This range therefore seems to give a fairly good estimate of the normal amplitude distribution.

In the diagnosis of neurogenic disorders the occurrence of fibrillation potentials is an important finding. These potentials usually have a short duration and a low amplitude (cf. Buchthal & Rosenfalck, 1966). Potentials of short duration occur also in normal laryngeal muscles and a special study of potentials with a duration $<2 \text{ ms}$ was made in the present material. Eighteen potentials were found comprising 5.3% of the total number. Their amplitudes were usually in the range $100\text{--}300 \mu\text{V}$.

but potentials with a much higher amplitude were also found. In Fig. 3 the amplitudes of all potentials $<2 \text{ ms}$ have been plotted. As will be seen, the distribution of these potentials shows a resemblance to that of the amplitudes of all potentials.

DISCUSSION

Earlier investigations have pointed to the difficulty of recording and measuring single motor unit potentials in human laryngeal muscles owing to their sustained activity (Weddell et al. 1944) which causes baseline disturbances and prevents a distinction to be made of the onset and end of a motor unit potential. Such sustained activity was an almost regular observation in the present investigations, but the difficulties in identifying action potentials could usually be reduced through minute alterations in the position of the needle electrode whereby motor unit potentials with less background disturbance could be recorded.

In electromyographical recordings from different points in the same muscle the duration, amplitude and shape of the action potentials may vary (cf. Buchthal & Clemmensen 1941; Petersén & Kugelberg, 1949; Buchthal et al. 1954). In this investigation recordings were made with the tip of the needle in many different positions to sample at random potentials from different sites within the muscle. Care was taken to identify about 20 motor unit potentials in each muscle to obtain a statistically significant number of potentials for analysis.

The recording procedure used enabled a study to be made of several motor unit potentials during repeated respiratory periods. Thus, there were motor units with constant discharge frequency and others which modulated their frequency with the respiration. Some motor units had a maximal discharge frequency during the inspiratory phase, others during the expiratory. The presence of inspiratory and expiratory motor units accords with findings made during recording of motor unit

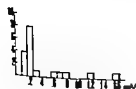


Fig. 3 Amplitude distribution of action potentials of duration 2 ms or less recorded from normal cricothyroid muscle. Ordinate: number of potentials.

potentials in the cricothyroid muscle of the cat (Rudomin 1966) and vocal muscle of man (Knutsson et al. 1969). In studies of the total electromyographic activity during respiration an increase of the activity has been observed both during the expiratory (Fink et al. 1956) and during the inspiratory phases (Faaborg-Andersen 1957).

In the present investigation the mean duration of the motor unit potentials was found to be 3.99 ± 1.40 ms (S.D. n 338), while in the only investigation earlier performed the mean duration was 5.3 ± 0.1 ms (S.E. n 201 Faaborg-Andersen 1957). The mean duration of motor unit potentials as recorded with different types of electrodes varies (Petersén & Kugelberg 1949). The difference in mean duration found in the present investigation and in that of Faaborg-Andersen is, however, not possible to explain in this way. Similar types of electrodes were used in both investigations the only difference being the outer diameter of the cannula. Such a difference in size of concentric needle electrodes does not affect the duration of recorded action potentials (Buchthal et al. 1954). However the difference in mean age may at least in part explain the difference in mean duration, since the mean duration of the motor unit potentials in a muscle decreases with increase of age (Petersén & Kugelberg 1949; Sacco et al. 1962). Thus, the mean age of the subjects in the present study was 31 years and of those studied by Faaborg-Andersen between 50 and 56 years. By way of comparison it may be stated that the mean duration in the vocal muscle in age groups 20-30 and 40-60 years was 3.43 ms and 4.21 ms, respectively. Also the difference in sampling of potentials may explain part of the difference in mean value of the duration. In the present investigation a sampling of motor unit potentials from many sites were made while in the investigations of Faaborg-Andersen only few potentials from a single place within each muscle were recorded.

The mean duration of the motor unit potentials in the vocal muscle was 3.76 ± 1.11 ms

(n 469; Knutsson et al. 1969). The difference between the values found in the vocal and in the cricothyroid muscle is small but statistically significant. Compared with the mean duration of potentials in limb muscles the durations of the potentials in the cricothyroid muscle are considerably shorter and could be placed in the same group as the vocal muscles, the extraocular muscles and the facial muscles.

When explaining differences in duration of motor unit potentials the longitudinal spread of the end plates of the motor unit and the consequent variation in distance which must be travelled by the muscle spikes before reaching the recording electrode has been shown to be an essential factor (Buchthal et al. 1955). In the cricothyroid muscle the motor end plates are localized to a narrow band in the middle of the muscle comprising about 15% of its length (Faaborg-Andersen, 1957). The total length of the cricothyroid muscle is 15 mm and the temporal dispersion of the muscle fiber spikes when arriving at the recording electrode with a velocity of about 4 m/sec must be comparatively small. This small temporal dispersion explains the short durations of the motor unit potentials found in the cricothyroid muscle.

The incidence of polyphasic potentials varies in different muscles. Thus, in normal limb muscles 2-4% of the potentials have more than 4 phases. In the facial muscles their frequency is still higher (Petersén & Kugelberg 1949) whereas no polyphasic potentials have been found in normal extraocular muscles (Björk & Kugelberg 1953). In single vocal muscle polyphasic potentials are present in up to 15% whereas in normal cricothyroid muscle up to 4%.

The present investigation reports data which constitute the main elements of the normal electromyographic pattern in the cricothyroid muscle and should provide a basis for a more complete electromyographical evaluation of laryngeal functions than previously possible. Thus, it may be used in motor disturbances of the vocal cord. In a series of electromyographical examinations (Haglund et al., 1970,

1971) it has been shown that a partial paresis of the recurrent nerve may exist although the laryngoscopic examination reveals a normal motility of the vocal cords, thus indicating that conventional methods cannot always reveal a partial paresis. In a subsequent paper examples of the deviations from normal electromyographic pattern in the cricothyroid muscle in some different types of lesions will be given.

ZUSAMMENFASSUNG

An 13 gesunden Menschen wurde das elektromyographische Muster von 17 cricothyroidalen Muskeln untersucht. Die EMG-Untersuchungen wurden mit einer konzentrischen Nadelelektrode durchgeführt. Bei ruhiger Atmung war bei der Mehrzahl der Muskeln eine gleichzeitige Aktivität während der Phasen der Respiration vorhanden. Einige motorische Einheiten entlockten sich mit gleichmäßiger Frequenz, wogegen bei anderen die Entladungsfrequenz je nach der Respirationsphase wechselte. Einige zeigten eine Zunahme der Aktivität während der Inspiration, andere dagegen bei der Expiration. Etwa 20 Potentiale der motorischen Einheiten wurden von jedem Muskel erhalten. Die Mehrzahl der Potentiale waren di- oder triphasisch. Polyphasische Potentiale erhielten wir bei 1% der gesamten Anzahl der Potentiale. Die mittlere Dauer der Potentiale war $3,99 \pm 1,40$ ms ($n=338$). Das Vorzeichen des Potential < 2 ms betrug im ganzen Material 5% und variierte in den individuellen Muskeln zwischen 0-16%. Die mittlere Amplitude war $394 \mu V$ ($n=338$). Das sechste und neuntzigste Perzentil betrug $150 \mu V$ bzw. $800 \mu V$. Die Maximalamplitude des Interferenzmusters lag gewöhnlich zwischen 1200 und 2000 μV .

REFERENCES

Björk, A. & Kugelberg, E. 1953 Motor unit activity in human extraocular muscles. *Electroenceph Clin Neurophysiol* 5 271

- Buchthal, F. & Clemmensen, S. 1941 On the differentiation of muscle atrophy by electromyography. *Acta Paediatr Scand* 16 143
- Buchthal, F., Guild, C. & Rosenfalck, P. 1954 Action potential parameters in normal human muscle and their dependence on physical variables. *Acta Physiol Scand* 32 200.
- 1955 Innervation zone and propagation velocity in human muscle. *Acta Physiol Scand* 35 174
- Buchthal, F. & Rosenfalck, P. 1966 Spontaneous electrical activity of human muscle. *Electroenceph Clin Neurophysiol* 20 321
- Fuhsborg-Andersen, A. 1957 Electromyographic investigations of intrinsic laryngeal muscles in humans. *Acta Physiol Scand* 41 Suppl. 140.
- Fink, B., Basak, M. & Epanchin, V. 1956 The mechanism of opening of the human larynx. *Laryngoscope* 66 410.
- Haglund, S., Knutsson, E., Mårtensson, A. 1972. An electromyographic analysis of idiopathic vocal cord paresis. *Acta Otolaryng (Stockh.)* 74 265
- Haglund, S., Knutsson, E., Mårtensson, A. & Mårtensson, B. 1970 Electromyography in vocal-cord paresis. *Acta Otolaryng (Stockh.)* 85 263
- Knutsson, E., Mårtensson, A. & Mårtensson, B. 1969 The normal electromyogram in human vocal muscles. *Acta Otolaryng (Stockh.)* 68 526.
- Petersén, I. & Kugelberg, E. 1949 Duration and form of action potentials in the normal human muscle. *J Neurol Neurosurg Psychiatr* 12 124
- Rodomin, P. 1966. The electrical activity of the cricothyroid muscles of the cat. *Arch Int Physiol* 74 135
- Sacco, G., Buchthal, F. & Rosenfalck, P. 1962. Motor unit potentials at different ages. *Arch Neurol* 6 366.
- Weddell, G., Feinstein, B. & Pattie, R. E. 1944 The electrical activity of voluntary muscle in man under normal and pathological conditions. *Brain* 67 178.

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ECHO GLOTTOGRAPHY

Ultrasonic Recording of Vocal Fold Vibrations in Preparations of Human Larynges

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(Received April 11 1972)

Abstract Except for the entirely subjective stroboscopy there is no generally accepted clinical method for continuous recording of vocal fold vibrations. In laryngology and phoniatrics it is therefore usually difficult to assess vocal fold function objectively. Echoglottoigraphy however an ultrasonic pulse-echo method for the study of the vibrating vocal folds, meets many of the necessary requirements.

For the recording of vocal fold vibrations a special, highly sensitive ultrasound reflectoscope has been constructed, allowing the use of transducers with different ultrasound frequencies. A pulse repetition frequency of 10 kHz was chosen, sufficient for continuous recording of fast vibratory movements, yet giving an adequate recording depth. In initial experiments an ultrasound frequency of 4 MHz was shown to penetrate even ossified cartilages, this frequency being high enough for high longitudinal and lateral resolution of the ultrasound beam.

As a basis for the development of an echoglotto-graphic method the ultrasound echoes from laryngeal specimens have been recorded. Echoes from the free margin of the vocal folds could be demonstrated and unequivocally identified. Vibrations in these same structures gave rise to characteristic curves, resembling glottograms recorded with other less convenient methods.

The vibratory pattern of the human vocal folds during phonation is of great interest in laryngeal physiology as well as in clinical laryngology. Because of their high frequency the vibrations of the folds cannot be visualized

directly but during recent decades several methods have come into use to overcome this obstacle. However since all of these more or less indirect methods have certain drawbacks, none of them has been generally adopted in clinical routine work at departments of laryngology.

The commonest clinical method is synchro-stroboscopy described in detail by Schönblin (1960). It provides no objective recording but is entirely dependent on the investigator's subjective impression of slow motion which appears when the human eye observes very short flashed images from consecutive vibratory cycles, and no continuous recording of the vibrations can be made. A great deal of research work on laryngeal physiology has been done with high speed film technique, first described by Farnsworth (1940) but in spite of isolated reports to the contrary (Teter et al, 1969), it must be said to be too expensive and too space and time-consuming for use in clinical routine. Besides, the employment of a fixed laryngeal mirror as part of this method demands a great deal of cooperation from the subject.

Untrained patients, who in addition, may present pharynx and larynx with narrow anatomy are therefore difficult to photograph. A comparatively easy way to record the

The investigation was supported in part by grants from Swedish Medical Research Council (Project K70-14X 3021-01A) and Swedish Board for Technical Development (Project 70-1364 U1033b)

continuous variations of the glottal area during phonation is by photoelectric glottography (Sonnesson, 1960). By this method the intensity variations of a transilluminated light beam directed through the glottis are recorded and visualized as a curve on a cathode ray oscilloscope. However the method does not allow conclusions concerning the vibratory movements of one single vocal fold, and some authors have pointed out that it yields insufficient information about certain parts of the vibratory cycle (Köster & Smith, 1970).

In electroglottography described by Fabre (1957) the impedance variations in the tissues of the neck are recorded during phonation. Because of variations in tissue impedance not dependent on vocal fold movements, this method involves a considerable risk of artefacts interfering with recording (Frøkjær Jensen, 1969).

In studies of laryngeal function many investigators have used acoustic analyses of the emitted voice, such as sound spectrography (Nessel, 1962; Yanagihara, 1962, 1967) or inverse filtering (Fant, 1961). In addition to other disadvantages of these methods in clinical work, only limited conclusions can be drawn from them as regards the vibratory pattern of each vocal fold.

It is obvious from these introductory remarks that an ultrasonic method for recording of vocal fold vibrations would be very convenient in clinical laryngology and phoniatrics, and it would also be very comfortable for the patient. The recording would be continuous, direct and objective—a combination not possible to achieve by any other single method. Besides, it would be possible to develop the method by using two transducers covering both vocal folds simultaneously.

The first to present an ultrasonic recording of a vibrating vocal fold was probably Mensch in 1964. Unfortunately his traces do not allow any analysis of the vibratory movements involved. In two almost identical papers Kitamura et al. (1968) have published very interesting recordings, and their forthcoming

more detailed report is eagerly looked forward to. Hertz et al. (1970) have presented a preliminary report on vocal fold recordings, prepared at our laboratory with an apparatus originally devised for echocardiography. This type of apparatus has a pulse repetition frequency of 1 000 Hz, which is not high enough to produce clinically usable recordings of vocal fold vibrations with a frequency of 100 Hz or more.

Therefore an ultrasonic reflectoscope has been constructed with a pulse repetition frequency of 10 000 Hz, which has proved to give satisfactory recordings of vocal fold vibrations. An outline of this new apparatus is given below. Technical details will be given in a special report to be published later.

PHYSICAL PROPERTIES OF ULTRASOUND

Before the technical data and the application of the ultrasonic echo method for studying the vibrating vocal folds are described, a very short description of the physical properties of ultrasound will be presented below, and illustrated in Fig. 1. For more detailed information the reader is referred to Hertz (1967).

Reflection

When a plane acoustic wave travelling in a medium impinges upon a boundary of a second medium, part of the wave is reflected into the first medium. The reflected sound intensity I resulting from a sound beam of intensity I_0 , falling perpendicularly on a flat surface, is represented by

$$I = I_0 \left(\frac{\rho_1 v_1 - \rho_2 v_2}{\rho_1 v_1 + \rho_2 v_2} \right)^2$$

where ρ and v are densities and sound velocities in two media on each side of a reflecting boundary (see Fig. 1 a). As the larynx is filled with air having an acoustic impedance, ρv which is much smaller than the surrounding tissues, strong reflection from the boundary vocal fold vs. air is to be expected.

Absorption

The absorption of ultrasound is considerable in biological tissue. If a parallel ultrasound beam travels through a certain medium the intensity of the beam decreases along the beam according to the relation

$$I = I_0 e^{-2kx}$$

where k is a constant depending on the biological substance, f the sound frequency and x the distance covered by the sound wave in the medium. This phenomenon is illustrated in Fig. 1 b. It appears from this that ultrasound of very high frequency is too much absorbed to be of practical use.

Diffraction

An ultrasound beam like any wave motion diverges because of diffraction. In the well-known Fraunhofer diffraction formula, $\sin \varphi = 0.61 \lambda / a$, the angle φ is the divergence angle, λ is the wavelength of sound and a the radius of a circular transducer. From this it is obvious that the use of a very high frequency (short wave length, λ) will result in a small diffraction angle and thus in a well-defined beam. The diffraction is illustrated in Fig. 1 c. It is important to notice that a small transducer may generate a poorly defined beam with a high divergence angle.

Angle error

If the reflecting surface is not perpendicular to the transmitter a great deal or all of the transmitted sound will not be reflected back to the transducer Fig. 1 d.

METHOD**Technical principles**

The ultrasonic pulse echo method is used for recording of vocal fold vibrations. The movements of the ultrasound echoes are recorded by the time motion (TM) technique well known from echocardiography Fig. 2 shows the principle of the time motion technique. A

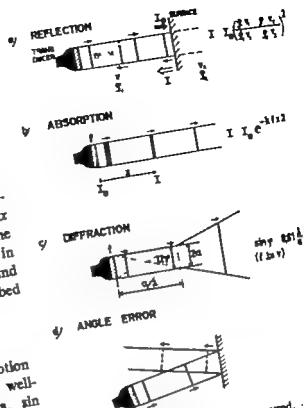


Fig. 1 Physical properties of ultrasound. — transmitted ultrasound pulse --- reflected ultrasound pulse. (a) Reflection of ultrasound beam towards a surface where g and v are densities and sound velocities in two media on each side of a reflecting boundary. (b) Absorption of ultrasound, where I is the constant depending on the biological tissue, f the sound frequency and x the distance covered by the sound. (c) Diffraction of an ultrasound beam. In the formula φ is the angle of divergence, λ the wavelength of sound in the medium and a the radius of a circular transducer. (d) Angle error. Loss of echo when beams do not strike reflecting surface at right angles.

saw tooth voltage generated from the ultrasound reflectoscope deflects the electron beam in a cathode ray tube (CRT) along the negative y-axis for every transmitted ultrasound pulse. At the same time the intensity of the electron beam in the CRT is modulated by the echo signal from the reflectoscope in such a way that it appears on the CRT screen only when an echo is received. If the time base (x-axis) of the cathode ray oscilloscope runs at a suitable slow deflection speed, the movements of the vocal fold echoes can immediately be observed on the screen. For

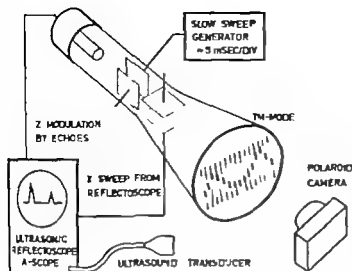


Fig 2 Principal diagram of the TM recording method in which the echo-glottographic recording of the vibrating vocal folds are directly displayed on the screen of a cathode ray tube and photographed with a Polaroid camera.

registration and filing the CRT-screen is photographed with a polaroid camera.

Reflectoscope

As mentioned above, commercial ultrasound reflectoscopes with a pulse repetition frequency of 50-1 000 Hz are useless because of the rapid vibratory movements of the vocal folds. Therefore the new ultrasound reflectoscope was built so as to yield a pulse repetition frequency of 10 000 Hz and was especially adapted for recording of the movements of the vocal folds. A block diagram of the apparatus is shown in Fig. 3.

The reflectoscope consists fundamentally of five blocks. The first block, a 10 kHz clock pulse generator controls the function of the following three blocks: sweep-generator, transmitter and depth compensation generator. The fifth block is the receiver unit with inputs from the transducer and the depth compensation generator and an output to the oscilloscope (TEK. 549) on which the TM-display is obtained. For control purposes, an A-scope display is achieved on a separate monitor oscilloscope.

The 10 kHz clock triggers the transmitter which generates 300 V pulses for the ultrasound transducer of only 10 μ sec duration.

This has the advantage of permitting the connection of transducers of different sizes and ultrasound frequency.

The 10 kHz clock also initiates the sweep generator which generates a saw tooth voltage each time an ultrasound pulse is transmitted. The start as well as the slope of the sweep can be adjusted. This makes it possible to select and magnify interesting echoes for TM display.

By the depth compensation generator losses due to ultrasound absorption in tissue can be neutralized within wide ranges.

The preamplifier of the receiver consists of five cascode amplifiers (Valley & Wallman, 1948) with overload protections. Cascode amplifiers have been chosen because of their excellent ability to amplify signals of very high frequency. The gain of this preamplifier is controlled by the depth compensation generator. Field effect transistors located near the cascode circuits adjust the gain of the receiver. They are operated by remote control which prevents the risk of undesirable oscillations.

For rectification, a new high-sensitive full-wave detector has been constructed. The rectified intensity modulation signal is transferred from a low impedance source via a coaxial cable to an amplifier placed immediately at

the cathode ray oscilloscope. This eliminates signal losses in the coaxial cable.

For TM representation of the vocal fold movements a storage oscilloscope is used. Because of the very high writing speed necessary a Tektronix 549 storage oscilloscope had to be chosen. The auto erase function of this CRO also makes it possible to discard less interesting echolotograms, which saves time and film costs.

Depending on the fundamental frequency of the vocal fold vibrations (100–200 Hz) the time base of the oscilloscope is used at a deflection speed of 1–10 msec/div.

The small size of the vibratory movements of the vocal folds puts the apparatus capacity for resolution to a severe test. The present equipment has experimentally been shown to measure correctly movements of 0.1 mm and less.

As the pulse repetition frequency is as high as 10 000 Hz, the silent period between two consecutive pulses has a duration of only 100 μ sec. This is the time in which returning echoes must be received. As the sound velocity in the tissues is about 1 500 m/sec, the maximum recording depth is calculated at somewhat more than 7 cm. At higher pulse repetition frequencies the recording depth decreases and there is a risk of receiving ghost echoes from earlier pulses.

Transducer

To be able to resolve the complex movements of the vocal folds, a very narrow ultrasonic beam must be used. Therefore the crystal beam must be selected with great care. A large crystal produces too wide a beam, a small crystal a beam of increasing angle of divergence. These problems are discussed extensively by Edler (1961). As one solution, ultrasound of as high a frequency as possible has been used, the frequency being limited by the sound absorption. Transducers with the frequencies 2, 4 and 6 MHz have been tested.

The transducer crystal is made of a ceramic material which converts an electrical voltage

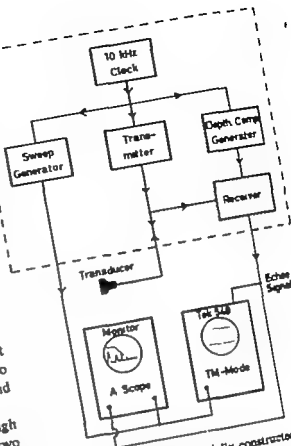


Fig 3 Block diagram of especially constructed refectoscope for echolotography including a monitor oscilloscope for A-scope, and a storage oscilloscope for TM-display

into sound pressure (pulse transmitter), and sound pressure to an electrical voltage (echo receiver). The material in the transducer (Brush Clevite PZT 5A), a modified lead zirconate titanate strikes a mean between transmuting efficiency and receiving sensitivity for the transducer together with a relatively high mechanical damping. To prevent the transducer from "ringing" it is necessary to mount the ceramic bowl or dish on a backing material which will increase the damping of the disc appreciably. This backing material consists of fine tungsten powder imbedded in Arakdite resin which efficiently scatters and absorbs the sound transmitted into it. The amount of tungsten powder is chosen in such a way that the acoustic impedance of the material is about the same as that required by the transducer to reach optimum damping.

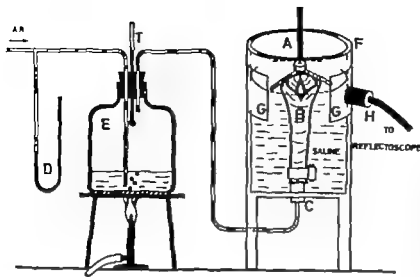


Fig 4 Arrangement of experiments. Moist and heated air passes through the trachea of a laryngeal preparation suspended in a saline-filled vessel with windows for the transducer.

With this transducer and external electrical damping it is possible to achieve sound pulses as short as 1 μ sec at 4 MHz.

Arrangement of experiments

In preliminary experiments ultrasound penetration of human thyroid cartilage with varying degree of ossification was compared with roentgenograms.

The arrangement shown in Fig 4 was used for the principal part of our work. On a fork like stand (A) the upper part of a human larynx preparation (B) was fixed, so that the

trachea was suspended vertically and could be connected to a plastic tube (C) for humidified and heated air (E). The temperature and pressure of the air could be measured by a thermometer (T) and a water-manometer (D) respectively. At certain air pressures and by manipulation of the laryngeal cartilages the vocal folds of the larynx preparation could be made to vibrate, emitting sounds of different pitch and quality. The larynx preparation was inserted into a circular perspex tank (F) with physiological saline or water. Windows (G) were cut in the tank and covered with a thin rubber membrane for application of the ultrasound transducer (H).

The experimental arrangements were made according to the principles developed by van den Berg et al. (1959).

RESULT

The ability of ultrasound to penetrate cartilage with varying degree of ossification was studied by measuring the amplitude attenuation at different frequencies. The results are shown in Table I. Measurements were made at the anterior edge and at the centre of four equally thick thyroid laminae varying in age and sex. The attenuation proved to be in good agree-

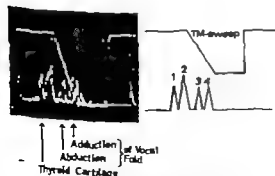


Fig 5 Ultrasound registration of manipulations with a vocal fold. A-scope. Double exposure with representation of the echoes from the same vocal fold in abducted and adducted position respectively. 1 External lamina, 2, internal lamina of thyroid cartilage; 3 vocal fold in abduction; 4 vocal fold in adduction.

Table 1 Attenuation of ultrasound intensity due to absorption in thyroid laminae (measured in dB = -10 log I₀/I)

Age; Sex	83/male		43/male		73/female		63/female	
	Anterior edge (-dB)	Centre (-dB)	Anterior edge (-dB)	Centre (-dB)	Anterior edge (-dB)	Centre (-dB)	Anterior edge (-dB)	Centre (-dB)
Transducer frequency MHz								
2	30	6	36	18	10	4	23	6
4	36	16	40	19	20	6	26	10
6		26	21	25	11	29	16	

Very high absorption, not measurable with the technique used.

ment with the ossification as it appeared on an X ray from the cartilages. As expected from textbook information about the progression of larynx ossification due to age and sex (e.g. Lanz & Wachsmuth 1955) absorption was shown to be least in the central part of the thyroid cartilage.

The human vocal folds are comparatively small structures: the size of their lateral excursion during vibration is often less than 1 mm. Therefore the best possible resolution is demanded from the ultrasonic apparatus. This can be achieved with ultrasound of high frequency. But, as pointed out earlier, the higher the frequency the greater the absorption, and there is a frequency limit when it becomes impossible to penetrate the tissues under investigation.

In our experiments the sound beam was in some cases practically entirely absorbed by the thyroid cartilage at a frequency of 6 MHz. The ultrasound frequency of 4 MHz was found to be the highest possible for experiments with larynx preparations, and the present work was carried out mainly with a transducer of this frequency.

At the beginning the experiments with

larynx preparations were carried out without vibrations of the vocal folds. An echo representing the inner and outer surface of the thyroid laminae of the preparation and another representing the free margin of the vocal fold were demonstrated on the monitor oscilloscope (A mode). This latter echo was unequivocally identified by the insertion of a thin metal needle under the marginal incision of the fold, which resulted in a change of the previously demonstrated echo. Further the echo could be identified by a good correspondence between the distance displayed on the CRT and the actual distance from the transducer to the free edge of the investigated vocal fold.

Manipulation of the fold, resulting in adduction and abduction movements, were followed as corresponding movements of the previously identified echo on the monitor whereas the remaining echoes did not move (Fig. 5). These slow movements of high amplitude were also demonstrated by TM recording as an undulating curve (Fig. 6).

When the vocal folds were made to vibrate by application of the moist and heated air stream, the excursions of the echo on the

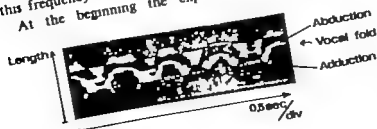


Fig. 6 Ultrasound registration of vocal fold movements with a vocal fold. TM-diagram. The slow ab- and adductory movements of the vocal fold are represented by an undulating curve.

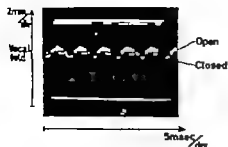


Fig. 7 Echoglotogram of vocal fold vibrating at 110 Hz. Note similarity of curves with the vibratory pattern well-known from high-speed films and photostereography. The closed, opening- and closing phases can clearly be distinguished.

monitor were of course too fast for the naked eye to follow and could only be perceived as a blurring of the vocal fold echo. By TM display however it was clearly possible to get distinct curves from the vibrating folds (Fig. 7). As seen from the figure, the repetition frequency of about 10 kHz was sufficient to give a continuous representation of vocal folds, vibrating at about 110 Hz.

DISCUSSION

Beach & Kelsey (1969) seem to be the only investigators who have correlated an ultrasonic

vocal cord recording with another sort of recording. They compared ultrasound Doppler signals from the region of the vocal folds with the actual vibratory phase as it appeared on the frames of a high-speed film. Velocities and displacements calculated from the Doppler signal did not always correlate well with the motions of the folds and the Doppler monitoring system proved unsatisfactory. It remains to be seen if this conclusion is valid also for the pulse echo method used in this paper. However we do not find this very probable in view of the data presented in this report. The complex vibratory movements of the vocal folds with continuously changing surfaces act as sources of multiple echoes which merge into a single Doppler signal. Its integrated velocity curve is not correlated with the motion of any actual part of the vocal fold surface. These problems do not arise with the present method as multiple echoes are not integrated but displayed separated in time. This means that the distance to the echo-emitting structure can be accurately determined and that the most relevant echo can be distinguished.

Unlike earlier authors, we have not confined ourselves to obtaining curves from out-

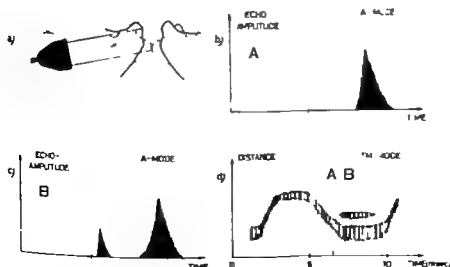


Fig. 8 a-d. Two diagrammatical frontal sections, A and B showing the complex and changing vocal fold surface during vibration (a), sometimes causing mul-

tiples and irregular echoes displayed in A-mode (b and c) as well as in TM-mode (d).

4 MHz ultraschallmässig durchzudringen. Diese Frequenz gestattet eine gute Auflösung bei der Aufzeichnung von sowohl Entfernungs- als auch Grössenschwankungen eines Objekts.

Als Ausgangspunkt für die weitere Entwicklung einer echoglottografischen Methode sind mit Ultraschall gewonnenen Aufzeichnungen von menschlichen Kehlkopfpräparaten studiert worden. Hierbei wurden Aufnahmen der freien Stimmbandanteile erzielt, deren Umfang nurzeigend feststand. Beim Anblasen und Schwingen der Stimmbänder ergaben sich Aufzeichnungen von charakteristischen Kurven, deren Form im Prinzip mit Glottogrammen von anderen, unstrahlischen Methoden übereinstimmte.

REFERENCES

- Baker D 1970. Pulsed ultrasonic Doppler blood-flow sensing. *IEEE Trans on Sonics and Ultrasonics* **SU 17** 3 170.
- Beach, J L & Kelsey C. A. 1969 Ultrasonic doppler monitoring of vocal-fold velocity and displacement. *J Acoust Soc Amer* **46** 1045
- Berg, J. van den et al. 1959 Results of experiments with human larynxes. *Pract Otorhinolaryng* (Basel) **21** 425
- Edler I. 1961. Ultrasound cardiography *Acta Med Scand*, Suppl. 370
- Fabre, Ph. 1957 Un procédé électrique permettant l'inscription de l'acoustique glottique au cours de la phonation: glottographie de basse fréquence. *Prendres résumés. Bull Acad Nat Med (Par.)* **121** 66.
- Fett, G. 1961 A new anti-resonance circuit for inverse filtering. *STL-Quart Progr Stat Reports* **4** 1
- Fleming, D W 1940. High-speed motion pictures of the human vocal cords. *Bell Telephone Laboratories Record*, **18** 203
- Frisker Jensen, B. 1969 Construction and comparative tests of two different types of glottographs. Paper delivered at Scandinavian Otorhinological Congress Helsinki 1969 From Institute of Phonetics, University of Copenhagen.
- Hertz, C. H. 1967 Ultrasonic engineering in heart diagnosis. *Amer J Cardiol* **19** 6.
- Hertz, C. H. & Edler I. 1956. Die Registrierung von Herzwandbewegungen mit Hilfe des Ultraschall-Impulsverfahrens. *Acustica* **6** 361
- Hertz, C. H. Lindström, K. & Sonesson, B. 1970. Ultrasonic recording of the vibrating vocal folds. *Acta Otolaryng* (Stockh.) **69** 223
- Jeppsson, S. 1961 Echoencephalography *Acta Chi Scand Suppl.* 272
- Kihamura, T et al. 1968. Ultrasonoglogography *Jap Soc Ultrasonics in Medicine* **5** 30.
- 1968 Ultrasonoglogography A preliminary report. *Ibid* **40**
- Kitzling, P & Sonesson, B. 1967 Shape and shift of the laryngeal ventricle during phonation. *Acta Otolaryng* (Stockh.) **63** 479
- Krüger J P & Smith, S. 1970. Zur Interpretation elektrischer und photoelektrischer Glottogramme *Folia Phoniat* (Basel) **22** 92.
- Lanz, T von & Wachsmuth, W 1955 *Praktische Anatomie* **1** Band, 2. Teil, Hft. 287 Springer Verlag, Berlin, Göttingen, Heidelberg.
- Nessel, E. 1962. Über das Tonfrequenzspektrum der pathologisch veränderten Stimme. *Acta Otolaryng* (Stockh.), Suppl. 157
- Schönbühl, E. 1960. *Die Stroboskopie in der praktischen Laryngologie* Thieme Verlag, Stuttgart.
- Sonesson, B. 1960. On the anatomy and vibratory pattern of the human vocal folds. With special reference to a photo-electrical method for studying the vibratory movements. *Acta Otolaryng* (Stockh.), Suppl. 156
- Teter H L et al. 1969 High-speed photography of the larynx in a clinical setting. *Ann Otol* **78** 1227
- Valley G E & Wallman, H. 1948 Vacuum tube amplifiers. *Massachusetts Institute of Technology Radiation Laboratory Series*, Vol. 18 New York.
- Yanagihara, N 1962. Acoustic studies on hoarseness. *Otorhinolaryng Clinica* (Kyoto) **55** 357
- 1967 Significance of harmonic changes and noise components in hoarseness. *J Speech Res* **10** 531
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QUANTITATIVE MEASUREMENT OF DROOLING

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Abstract A method for quantitative measurement of drooling, utilizing the radioactive isotope ^{99m}Tc , is introduced. The aim of this method is to evaluate results of therapy against this state. Drool patients were given 100 μCi ^{99m}Tc intravenously. Saliva samples were repeatedly taken from the mouth for 9 hours and the isotope concentration was examined. The drooled saliva was collected in disposable bibs. From the measured radioactivity in the bibs and the saliva samples, the amount of drooled saliva could be calculated. The results show as regards saliva production in a normal population, great variations in amount of drooled saliva within as well as between individuals. With due precautions, the method is useful for quantitative measurement of drooling.

Every year between 200-300 children with cerebral palsy are born in Sweden. Among these about 10 (Enfors & Lundberg 1968) will have an embarrassing drooling. This causes much trouble to the child as well as to its parents or nursing personnel. Treatment against drooling has hitherto mainly been medical (Goode & Smith, 1970). A few cases surgically treated are described in the literature (Wilkie 1967, Enfors & Lundberg 1968, Laage-Hellman, 1969). The appraisal of the results after treatment has hitherto only been subjective, which makes a satisfactory comparison between different therapeutic results difficult. A method for quantitative measurement of the amount of drooled saliva therefore is desirable. As the amount of drooled saliva generally does not exceed 20 ml per hour, evaporation is suf-

ficiently important to preclude a simple weighing performance. A method which utilizes the radioactive isotope technetium (^{99m}Tc) is described in this study.

^{99m}Tc in the form of pertechnetate has in recent years been increasingly used in different studies of the function of the salivary glands (Abramson et al., 1969, Gates, 1970, Harden et al., 1969). Technetium was discovered in 1937 by Segré and was used clinically for the first time by McAfee et al., who in 1964 made the first brain scan. Technetium is found in the group VII a of the periodic system together with manganese and rhenium. The reason why technetium is so suitable for medical investigation is its physical properties. ^{99m}Tc is a daughter nuclide of molybdenum (^{99}Mo) has a physical half life of 6 hours and is a gamma-emitter of 140 keV. The deposition in the body is extremely small. The compound most frequently used is the peroxyanion TeO_4^{2-} also called pertechnetate. This ion is fairly stable, resembling iodide in its distribution in the human body. Both ions are concentrated in the salivary glands, iodide about 40 times and pertechnetate about 20 times the plasma values (Harden & Alexander 1967, Gates, 1970). One hour after intravenous injection of pertechnetate, 30% of the activity is found in the red cell fraction and 70% in the plasma. 75% of the plasma activity is protein-bound (McAfee et al., 1964). The radioactive daughter ^{99m}Tc arises from decay of ^{99}Tc . The radiation dosage of ^{99m}Tc

This study was supported by Grants from Folke Bernadotte Stiftelsen and First of May Flower Annual Campaign for Children's Health

is negligible, because of a very long physical half-life (2×10^5 years). Excretion of ^{99m}Tc is quickest within the first 24 hours after administration, and mainly in the urine (Harper et al., 1965).

One advantage of pertechnetate compared with iodide is that the activity used for scintigraphy results in less radiation dose to the investigated person. A thyroid scintigram with the usual activity of 50 μCi ^{131}I results in an irradiation against the gland of about 100 rad. If ^{99m}Tc is used instead, in the ordinary dose of 1 mCi, the thyroid gland is irradiated 1 000 times less, i.e. 100 mrad (Harper et al. 1965). To prepare a salivary gland scintigram, a sufficient dose would be 100–200 μCi ^{99m}Tc , i.e. an irradiation in the salivary glands of 4–8 mrad (Grünberg & Börner 1966).

Within 15 minutes of intravenous administration of pertechnetate, the isotope is found in the saliva. The most likely site for the concentration of the isotope is the ductal epithelium (Abramson et al., 1969). After intravenous injection of pertechnetate, the isotope could be demonstrated in the great, paired, salivary glands as well as in the acinar glands in the oral mucosa. It has long been well known what happens to iodide in the body after intravenous or peroral administration. Experiments with pertechnetate indicate the same distribution. Harden & Alexander (1967) compared the clearance of iodide and pertechnetate in the parotid gland. They found that increased secretion of saliva resulted in higher clearance of both iodide and pertechnetate, the relation, however being constant 2:1. An increased secretion rate resulted in lower viscosity and lower concentration of technetium in the saliva. Harden considers that pertechnetate and iodide share a common transport mechanism.

The aim of the present investigations was to measure the amount of drooled saliva. First of all, the best suited conditions for the investigation had to be determined. The criterion was that the absolute change of concentration had to be small within the actual time interval. In

the first series, changes in salivary concentration of the isotope after intravenous injection were studied. From these results a second series was planned where the amount of drooled saliva was determined by measuring the radioactivity of the bibs where drooled saliva was collected and of saliva samples taken during the same period.

MATERIAL AND METHOD

Series No 1

The series comprised 14 persons with brain damage mainly in the form of cerebral palsy. The age distribution was 9–23 years, 8 women and 6 men. Every patient was investigated twice. After intravenous injection of 100 μCi ^{99m}Tc , saliva was taken from the frontal part of the mouth every 15 min during the first hour and then after 1.5, 2, 3 and 4 hours. The samples were placed in airtight plastic cylinders which were examined in respect of radioactivity. The measuring device was a scintillation spectrometer type Picker Autowell with a 3-inch well crystal of sodium iodide. From the injected isotope solution a sample was withdrawn as a reference before the start of the investigation. The reference solution was used to show the decay of the isotope. The reference solution and the saliva samples were measured in identical geometry. The relation between radioactivity in saliva samples at different times and the reference solution was expressed in % of a given amount of radioisotope per gram of saliva. The mean figures from this series are shown in Table 1. Since the amount of radioisotope given was 100 μCi , the salivary isotope concentration in g of saliva corresponds to the same figures expressed as $\mu\text{Ci/g}$.

Discussion

The aim of this series of investigations was first to find out how the salivary concentration of ^{99m}Tc varied after intravenous administration. From the natural relation to the plasma concentration curve, one might expect that the isotope concentration in saliva would have a quick initial decrease followed by a slower secondary phase, which is mostly the result of a continuous excretion. The second purpose of this survey was to estimate the time interval suitable for the quantitative measurement of the amount of drooled saliva. In order to find an interval with a low relative variation, the observed mean values were compared to a model of regression representing

a constant, relative reduction of the salivary isotope concentration.

The mathematical model used is then assumed to be

$$Y = a e^{bt} + c \quad t > 0 \quad a > 0 \quad b > 0 \quad \text{where}$$

Y represents the mean of salivary concentration of ^{99m}Tc and t the time after injection. a and b are unknown constants and c is the base of natural logarithms. The mathematical model was fitted to the values in Table I.

The results are shown in Fig. 1. The fit of the mathematical model is not good in the first period. The relative reduction in salivary isotope concentration expressed in % per gram of saliva is greater at the beginning than at the end of the period of investigation. With these results as a background it is evident that quantitative measuring of drooled saliva should not start until 120 min after intravenous injection of ^{99m}Tc . One more reason for choosing this period is the greater individual variations of the concentration values seen at the beginning of the investigation. The series also shows that the process can be regarded as approximately linear from this point of time which simplifies the calculations made in series no. 2. The effects of the change caused by the continuous excretion of the isotope is, however, due to the prolonged observation period, still as great, and must be borne in mind in the evaluation of the results.

Series No. 2

Quantitative measurement of drooling was made in 12 persons with brain damage mainly in the form of cerebral palsy. Their intellect varied from normal to idiosyncrasy. The age distribution was 6–26 years in 11 persons and the last one was 46 years old. There was an equal distribution between the sexes. The investigations were made mainly in the institution where the patients lived in order to minimize changes in the daily routine and environment.

Discussion

Based on the results from series no. 1 the measurements were made 120–540 min after intravenous injection of $100 \mu\text{Ci}$ of ^{99m}Tc . The drooled saliva was collected in disposable bibs

Table I Observed values of ^{99m}Tc -concentration in saliva

14 patients investigated twice (224 samples). Given dose $100 \mu\text{Ci}$

Time after injection (minutes)	Concentration (%/g or $\mu\text{Ci/g}$)		
	Max.	Min.	Mean
15	0.91	0.17	0.35
30	0.54	0.15	0.29
45	0.67	0.10	0.26
60	0.52	0.08	0.23
90	0.51	0.09	0.20
120	0.33	0.10	0.18
180	0.28	0.08	0.15
240	0.27	0.07	0.12

40 x 50 cm made of one layer of a plastic sheet between two layers of cellulose wadding (Mölnlycke art. 720500).

Each bib was placed around the patient's neck for half an hour and was afterwards put into a plastic bag, which was then measured in respect of radioactivity. The measuring device consisted of a Packard Model 3002 Tri-Carb scintillation spectrometer and an Armac Model 440 scintillation detector. Each time the bib was changed a sample of saliva was taken with a pipette from the frontal part of the mouth. The saliva was put into an airtight plastic cylinder and weighed whereafter the radioactivity was determined in a spectrometer type Picker Autowell with a 3-inch well crystal of sodium iodide. A reference solution was used to show the decay of the isotope. Identical measuring geometry between the reference solutions and the saliva samples as well as the bibs was arranged. The measured radioactivity per weight unit of saliva and of the bibs, was used to calculate the amount of drooled saliva.

RESULT

The calculated figures of drooled saliva in series no. 2 are shown in Table II. Each value is the mean of 14 measurements. Altogether 42 measurements of drooled saliva were made on every patient. Within one individual there could be a variation of 0.4–4.0 g drooled saliva

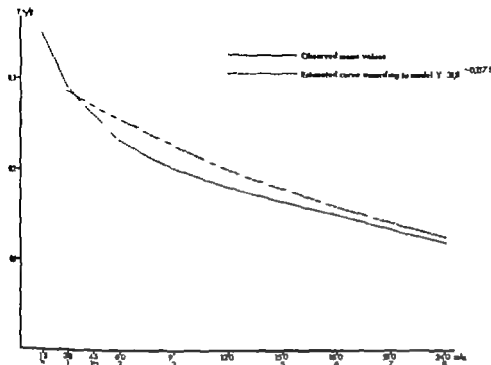


Fig. 1 Salivary concentration of ^{99m}Tc .

per 30 min. These variations existed independently of the investigations being made within a few hours or several weeks. Variations between individuals were found from 0.4–16.7 g drooled saliva per 30 min.

STATISTICAL ANALYSIS

1. Analysis of regression

On three different occasions 14 measurements were made. To reduce the errors of the method in analysing the results, the average salivary isotope concentration values at different times, could be calculated for each person from the equation of linear regression, which, based on the results of series no. 1 could be accepted. The model of the equation this time was $Y = a + b \cdot t$. The concentration values were then used for the calculation of drooled saliva, thereby reducing the errors by 10–20%.

2. Analysis of variance

The values of drooled saliva in g per 30 min were treated with analysis of variance to test

the variations between the individuals and the variations between the means for different investigations of the same individuals. Statistically significant differences were received for both sources of variation. The results are shown in Table III.

DISCUSSION

The present paper describes a method for the quantitative measurement of drooling by using the radioactive isotope ^{99m}Tc . The aim was to make it possible to evaluate results of therapy against drooling. In evaluating the method some errors ought to be discussed. (a) variations in salivary isotope concentration, (b) variations in amount of saliva produced, (c) variations in amount of drooled saliva, (d) risk of irradiation.

In an article from 1967 Harden shows that the salivary concentration of ^{99m}Tc diminishes when the velocity of secretion is higher. This means that if the saliva sample is taken during a phase with high secretion rate, the

of drooled saliva is overestimated. The opposite result is obtained if the sample is taken in a phase of low salivary secretion. Hence this error is randomized.

It is well known from the literature that there are great variations of saliva production in human beings (Becks & Wainwright, 1943; Gerke & Klemm, 1951; Schneyer 1956; Rauch 1959; Enfors, 1962). This fact is true both within the same person at varying times and between different persons. Even if it is not clear whether a drooling person drools a constant fraction of the total saliva produced it is possible that the great variations in amount of drooled saliva described are mainly explained by the variations in the total amount of saliva produced. From a methodological point of view these variations are embarrassing. We therefore have tried in this investigation to avoid factors influencing the total amount of saliva production. For this reason the measurements are made in the environment where the children live, trying to maintain as far as possible the daily routine including the regular meals, and with the ordinary nursing personnel present. The actual situation of investigation however must influence the saliva production too. With the knowledge of these variations it is evident that this measurement of drooling must be performed over a relatively long period and must be based on many samples. How great a number of samples, depends on the accuracy demanded in calculating the mean.

At the time of investigation the patient receives $100 \mu\text{Ci } ^{99m}\text{Tc}$ intravenously which gives an irradiation in the salivary glands of about 4

Table II Mean values of drooled saliva (g/30 min)
Each value is the mean of 14 measurements (304 samples)

Patients	Time of Investigation		
	I	II	III
K. A.	9.22	4.27	1.87
C. H.	3.16	2.52	3.08
B. J.	8.52	11.78	9.41
P. J.	2.07	3.49	3.62
B. K.	8.59	8.62	7.87
A. L.	6.12	1.79	3.86
B. O.	5.43	6.01	5.08
C. P.	1.59	1.17	2.60
J. P.	2.06	8.52	2.49
M. S.	5.01	2.95	3.53
L. S.	4.02	5.74	1.83
L. U.	7.11	8.45	11.53

mrads (Grünberg & Börner 1966). The calculated dose in the gonads in such a patient will be 1.2–1.6 mrad (Smith, 1965). As a comparison a conventional X-ray examination of the hypopharynx and oesophagus gives an irradiation of the gonads of about 20 mrad (Hammer Jacobsen, 1963). Thus the dose to the examined person in quantitative measuring of drooling is small.

Even if great changes of drooling in individuals with cerebral palsy can be observed by the nursing personnel, subjective observations of this kind cannot possibly be used to evaluate the results from different kinds of therapy given to a group of patients. The basis for correct evaluations must be a more objective method. If proper consideration is given to the above analysed errors and if a sufficient number of measurements are made, the method described in this paper is useful for quantitative measurements of drooled saliva.

Table III Analysis of variance on drooled saliva values (g/30 min)

Sources of variation	Degrees of freedom	Mean squares	F	Coefficient of variation (%)
Between individuals	11	271.86	5.21	44
Within individuals	26	51.43	6.99	34
Different times of investigation	468	7.80		54

ACKNOWLEDGEMENTS

The authors gratefully acknowledge the assistance in the statistical work of Professor Uno Zackrisson. Thanks is also expressed to the hospital physicists Agne Larsson and Bengt Roos for their interest and help in this investigation.

ZUSAMMENFASSUNG

Eine Methode für die quantitative Messung von giftigen Speichel wird vorgelegt. Die Methode benutzt den radioaktiven Isotop ^{99m}Tc Technetium und soll die Resultate der Therapie gegen Geffer schätzen. Geifernde Patienten bekommen $100\ \mu\text{Ci}\ ^{99m}\text{Tc}$ intravenös. Speichersproben wurden während 9 Stunden wiederholt dem Mund entnommen, worauf die Konzentration des Isotops gemessen wurde. Der giftige Speichel wurde in einem Gefferisatz gesammelt. Aus der gemessenen Radioaktivität des Gefferisatzes und der Speichersprobe konnte die Menge giftigen Speichels berechnet werden. Die Resultate zeigen, in Übereinstimmung mit dem Speichsekret einer normalen Population, grosse Unterschiede der Menge giftigen Speichels sowohl bei einer als auch unter den untersuchten Personen. Eine Variationsanalyse wird gemacht, und die Fehler werden diskutiert. Insgesamt ist die Methode brauchbar giftigen Speichel quantitativ zu messen.

REFERENCES

- Abramson, A. L., Levy I. M., Goodman, M. & Arzoo, J. N. 1969 Salivary gland scintiscanning with technetium 99m pertechnetate. *Laryngoscope* 79 1105.
- Beck, H. & Waterwright, W. W. 1943 Human saliva. XIII Rate of flow of resting saliva of healthy individuals. *J Dent Res* 22 391.
- Efors, B. 1962. The parotid and submandibular secretion in man. *Acta Otolaryng (Stockh)*, Suppl 172.
- Efors, B. & Lundberg, A. 1968. Behandling av hyperaktivitet hos cp-barn. *Läkartidningen* 65 4416.
- Gates, G. A. 1970. Current status of radiosialography in tumor diagnosis. *Trans Amer Acad Ophthalmol Otolaryng* 74 1183.
- Gerke, J. & Klemm, W. 1951 Beeinflussung der Speichelsekretion mit Kaugummi. *Deutsche Zahnärztliche Zeitschrift* 6 1181.
- Goode, R. L. & Smith, R. A. 1970. The surgical management of sialorhiza. *Laryngoscope* 80 1078.
- Grönberg, H. & Börner, W. 1966. Die ^{99m}Tc Pertechnetat Szintigraphie in der Diagnostik von Krankheiten der Kopfspeicheldrüsen. *Arch Klin Exp Ohr Nas Kehlkopfheilk* 187 714.
- Hansson Jacobson, E. 1963 Genetically significant radiation doses in diagnostic radiology. *Acta Radiol (Stockh)*, Suppl. 222.
- Harden, R. Mc G. & Alexander W. D. 1967 The relation between the clearance of iodide and pertechnetate in human parotid saliva and salivary flow rate. *Clin Sci* 33 425.
- Harden, R. Mc G. Alexander W. D. Shimmin, J. & Chisholm, D. 1969 A comparison between the gastric and salivary concentration of iodide, pertechnetate and bromide in man. *Gut*, 10 978.
- Harper P. V., Lathrop, K. A., Jimenez, F., Fink, R. & Gottschalk, A. 1965 Technetium 99m as a scanning agent. *Radiology* 85 101.
- Laage-Hellman, J.-E. 1969 Retroposition av gl. submandibularis' utforsking som behandling vid drooling. *Neurol Med* 82 1522.
- McAfee, J. G., Foeger, C. F., Stern, H. S., Wagner, H. N. Jr & Migha, T. 1964 Technetium 99m pertechnetate for brain scanning. *J Nucl Med* 5 811.
- Rauch, S. 1959 *Die Speicheldrüsen des Menschen*. G. Thieme Verlag, Stuttgart.
- Schöyner, L. H. 1956. Source of resting total mixed saliva of man. *J Appl Physiol* 9 79.
- Smith, E. M. 1965 Internal dose calculation for ^{99m}Tc . *J Nucl Med* 6 231.
- Willie, T. F. 1967 The problem of drooling in cerebral palsy: A surgical approach. *Canad J Surg* 10 60.
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International Otoneurological Symposium

Basel 1969 Editor: C. R. Pfeils (Basel)

Transtemporal Surgery of the Internal Auditory Canal by U. Fisch (Zürich)

viii + 39 p., 125 fig., 11 tab., 1970. SFr. 69.50/US\$ 19.50/DM 69.50/£ 7.68
ISBN 3-8055-0237-0

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ADAPTATION OF HORIZONTAL SEMICIRCULAR CANAL RESPONSES

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(Received August 28, 1972)

Abstract. The torsion pendulum model predicts semicircular canal responses when angular accelerative stimuli are relatively brief, but certain discrepancies appear when accelerations are sustained for a longer time. A recently proposed revision of the torsion pendulum model accounts for many of these discrepancies on the basis of a short-term adaptation process. The revised model was tested by comparing its predictions with the observed responses of subjects exposed to a series of prolonged accelerations. The model described these data fairly accurately although minor discrepancies remained, suggesting the existence of additional adaptation processes. A new method is described for measuring the adaptation process described by the model. This method is based on a single measurement that can be obtained with relative ease for either nystagmus or sensation responses.

A mathematical model can be of considerable value in the study of a sensory system. A model provides a general equation from which the responses can be predicted for any stimulus within a specified range. Further if the model is parsimonious and the system under consideration is reasonably simple the response can be completely specified by a small number of parameters.

The torsion pendulum model of the semicircular canal system (Van Egmond et al., 1949) is such a model. It permits specification of the response to any angular accelerative stimulus if the values of two parameters—the cupula long time constant (τ) and the response gain (K_d)—are known. The value of this model is widely recognized but its range of application is limited by the fact that it accurately describes responses only when the semicircular canal stimulus is relatively brief

Whenever an acceleration is prolonged beyond a few seconds, the observed response departs systematically from the model's prediction (Mowrer 1935 Ek et al., 1960 Guedry & Lauver 1961). A number of workers (Ek et al., 1960 Hood, 1961 Aschan & Bergstedt, 1965 Guedry 1965) have suggested that this discrepancy is not caused by an error in the model itself but rather by the intervention of response adaptation, for which the model does not account.

A more comprehensive model, which includes both the torsion pendulum model and an adaptation term has been proposed by Young & Oman (1969) and by Malcolm & Jones (1970). These authors assume that the observed semicircular canal response is proportional to the difference between the input signal from the canals and a "zero-velocity reference level," which exists somewhere within the nervous system. The response appears to adapt because this reference level shifts during sustained stimulation in the direction that reduces the difference between itself and the sensory input level. The authors further assume that the rate at which the reference level shifts is dependent upon the magnitude of this difference.

The revised model, if correct, considerably extends the range of accurate prediction. It has already proven successful in predicting responses to several types of prolonged accelerations (Young & Oman, 1969). The model is also parsimonious. In addition to the

parameters (τ_0 and $K_{\frac{1}{2}}^0$) mentioned previously the model includes only one other parameter—an adaptation time constant (τ)

Methods have been previously described for estimating the τ and $K_{\frac{1}{2}}^0$ parameters (Guedry et al. 1971a,b; Gilson et al. 1973) which meet practical requirements with respect to subject comfort and administration time. These methods are of course based on the use of brief stimuli in which the confounding effects of adaptation presumably do not occur. In this investigation an attempt is made to develop a method of estimating the adaptation parameter (τ) that also meets practical requirements for routine use.

This report describes two experiments. The first examines the ability of the revised model to describe responses to the particular type of stimulus under consideration and thus provides an additional test of the model's validity. The subjects in this experiment are tested under two conditions of mental alertness to determine if the level of alertness has any influence on the rate of adaptation. The second experiment evaluates a proposed method for estimating τ_0 .

EXPERIMENT 1

Procedure

2 U.S. Navy flight students served as subjects. All had passed U.S. Naval flight physical examinations without evidence of vestibular disorder. Each student was administered four semicircular canal stimuli with the horizontal canals in the plane of rotation. The stimulus in each case was an angular acceleration of 5 deg/sec^2 for 60 sec, followed immediately by a deceleration of the same magnitude and duration. Nystagmus was recorded during, and for 3 min after each presentation of the stimulus by the electrooculographic method. During two of the trials (one clockwise and the other counterclockwise) the subject was kept alert with mental arithmetic. During the other two trials (one clockwise and the other counterclockwise) he was unoccupied by any

task and was told to relax. The order of administration of trials was counterbalanced across subjects to avoid contamination of the results by serial effects.

Since it is known that dark adaptation affects the corneoretinal potential (Gonsky & Malcolm 1971) all subjects were completely dark-adapted before the experiment began and were maintained in red light during the 10 min rest periods between trials. Subjects were always in complete darkness while nystagmus was being recorded.

Results and discussion

The data shown in Fig. 1 represent the averaged nystagmus responses of all six subjects while they performed mental arithmetic. The responses of these subjects clearly show the phenomena attributable to adaptation viz., a response decline during constant acceleration, and a secondary response appearing after the stimulus had ceased. The torsion pendulum model predicts neither of these phenomena, but as shown in Fig. 1 the revised model provides a reasonably accurate description of both. Parametric values were selected by first setting τ at 16 sec, a value that has been found previously (Gilson et al. 1973), then choosing values of $K_{\frac{1}{2}}^0$ and τ_0 to obtain the best visual fit to the data. The optimum value of τ was 120 sec, which is similar to the values found by other investigators to provide the best fit with data from a variety of sources (Young & Oman 1969; Malcolm & Jones, 1970).

To the extent that the data match the theoretical curve, they confirm the adequacy of the revised model. The fit of the model is acceptable, although minor discrepancies exist, which may suggest the operation of other adaptation processes besides the one described by the model. One such process might be a progressive reduction of the response gain ($K_{\frac{1}{2}}^0$) during sustained acceleration. A preliminary effort at describing the data with a model that includes a progressive gain reduction has yielded a better fit than does the

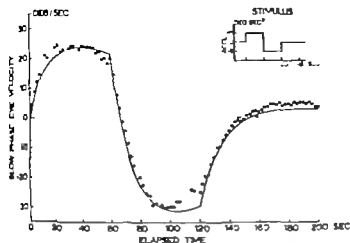


Fig 1 Nystagmus response to a triangular waveform stimulus when subjects were alert (a). Data for clockwise and counterclockwise rotation are combined: positive values indicate nystagmus slow phase velocity opposite the direction of rotation (6 subjects, 2 trials). Also shown is the revised model's prediction of the response with $\tau = 16$ sec and $\tau = 120$ sec. The response obtained while subjects were not alert is not shown: an accurate fit to those data was obtained by decreasing the value of K_{Σ}^0 without altering the values of τ or τ_0 .

prediction shown in Fig 1. However the improvement is rather slight for the stimulus used here. Experiments are underway using more prolonged stimuli that are designed to clearly demonstrate a progressive gain reduction, if in fact one exists.

During the trials in which subjects had no task to perform, the nystagmus responses were reduced, as expected from previous studies (Guedry & Lauver 1961, Collins & Guedry 1962). However this reduction (about 10%) was not as great as some investigators have found. It is quite possible that, since the subjects were student aviators undergoing a period of intense evaluation at the time of testing, they found the experimental situation too alerting and could not relax appreciably during the trials in which they had no task to perform. The small reduction that was found could be accounted for by substituting a lower response gain (K_{Σ}^0) into the prediction equation while keeping the value of τ unchanged. Thus, the adaptation rate did not appear to be influenced by the level of alertness in this study but there remains a possibility that such an effect might have been revealed if greater differences in alertness had been obtained.

EXPERIMENT 2

It is well known that the sensation of rotation exhibits a faster decline during prolonged rota-

tion than does nystagmus (Ek et al., 1960; Guedry & Lauver 1961). Young & Oman (1969) accounted for this phenomenon in their revised model by assuming that the adaptation rate is faster (i.e., τ is smaller) for sensations than it is for nystagmus. This assumption implies that adaptation occurs somewhat independently in these two response systems. If so, then any method for assessing adaptation should be applicable to both of them. Most investigators have preferred to observe nystagmus, methods that have been developed for measuring sensations are generally too laborious for routine use. However a recently reported technique for measuring subjective turning points (Guedry et al., 1971a) appears suitable for this purpose. Under this method, the subject is exposed in darkness to a series of stimuli like those used in the previous experiment. The sensation of a normal individual in this situation is similar in form to the nystagmus response shown in Fig 1, i.e., he first experiences acceleration in the proper direction then deceleration, then a momentary pause, and then commencement of rotation in the opposite direction. The subject's task is to signal at the moment he experiences this reversal in direction of rotation.¹

After the stimulation stops, the subject will experience deceleration again, and, if the stimulus is sufficiently long, he may experience another reversal in the direction of rotation (see Fig. 1). The reversal was not studied in this experiment.

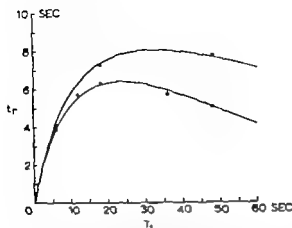


Fig. 2 t values for nystagmus (●) and for sensation (○) to stimuli of various durations (6 subjects \times 6 trials). Also shown is a description by Eq. 1 of nystagmus data with $\tau = 16$ sec and $\tau = 150$ sec, and a description of sensation data with $\tau = 16$ sec and $\tau = 50$ sec.

If t is the time interval from the end of acceleration to the sensation of reversal, T_1 is the duration of acceleration, τ is the time constant of the cupula response, and τ_c is the time constant of adaptation, then

$$t = \left(\frac{1}{1/\tau_c - 1/\tau} \right) \ln \left(\frac{2 - e^{-T_1/\tau}}{2 - e^{-T_1/\tau_c}} \right) \quad (\text{Eq. 1})$$

Thus, t depends upon the values of two parameters— τ and τ_c . When τ is known τ_c for sensation can be determined.

Eq. 1 can be used to determine τ for nystagmus as well, since nystagmus t can be easily found by measuring (on the nystagmus record) the time interval from the end of T_1 until the reversal in the direction of nystagmus.

In this experiment subjects were exposed to a series of prolonged stimuli and values for both sensation and nystagmus τ were determined.

Procedure

Six individuals (Naval officers and laboratory personnel) served as subjects in this experiment. All were free from obvious vestibular defects. Each subject was exposed in darkness to a series of triangular waveform stimuli in the

plane of the horizontal semicircular canals. The stimulus in each case was an angular acceleration of 5 deg/sec for a given duration followed immediately by a deceleration of the same duration and magnitude. The duration of the acceleration phase of the stimulus (T_1) was varied in seven steps ($T_1 = 6, 12, 18, 24, 36, 48$, and 60 sec) with each duration presented six times (three times in the clockwise direction and three times in the counterclockwise direction in alternating order). The order of administration of stimuli was randomized across subjects to avoid serial effects. Rest periods equal to approximately five times the stimulus duration were allowed between trials, and the test series was spread over four testing sessions to minimize subject fatigue.

On each trial nystagmus was recorded, and subjects were asked to press a button at the moment they experienced the reversal in the direction of rotation. The t for sensation was determined for each trial by measuring the interval from the end of T_1 until the push-button response and t for nystagmus was determined by measuring the interval from the end of T_1 until nystagmus reversal. From these data, the values of subjective τ_c and nystagmus τ were estimated.

Results and discussion

Eq. 1 specifically predicts that, as T_1 increases, the t interval will grow longer until it reaches a maximum and then it will begin to grow shorter. Furthermore, if the subjective response does in fact adapt faster than nystagmus, the t for sensation should reach a maximum sooner than t for nystagmus and then decline more rapidly. The data, shown in Fig. 2, confirm all of these predictions. With τ set at 16 sec, an acceptable fit with the nystagmus data was obtained when τ_c for nystagmus was 150 sec, and a fit with subjective data was obtained when τ_c for sensation was 50 sec. Both values are somewhat similar to those employed by Young & Orman (1969) to describe data from other sources. The accuracy of the prediction thus offers additional support for the

revised model, including the assumption that the adaptation rate for the sensation of rotation is faster than the adaptation rate for nystagmus.

The method used here for measuring adaptation rate appears to yield reliable results suitable for routine assessment of individual differences, since it utilizes a single response measurement (t) that can be obtained relatively easily for either nystagmus or sensation. Most subjects are able to make consistent judgments of the subjective reversal with only brief instruction. However it was found that each subject must fully understand that he is to respond *immediately* after experiencing the reversal. Otherwise spuriously long t values were obtained.

In this experiment the testing procedure was somewhat lengthy because the subjects were exposed to seven durations of stimulus in order to provide enough data for an adequate test of the model. In practice, of course, only one stimulus duration (for example, $T_1 = 60$ sec) need be presented to obtain an estimate of τ_n and the number of trials can be chosen to meet practical requirements. The method requires separate determination of the cupula time constant (τ_c), but this value can be estimated by methods described in a previous report (Gilson et al. 1973).

An accurate description of the adaptation process significantly expands the universe of prediction, permitting prediction of responses under circumstances in which adaptation occurs or on the other hand, permitting selection of vestibular testing situations that avoid the confounding effects of adaptation. The diagnostic significance of adaptation measurements in detecting vestibular disorders remains to be determined by evaluation with clinical case material, but for some applications, such as evaluation of flight personnel, assessment of adaptive ability has a certain face validity since aviators must frequently cope with prolonged accelerations during aircraft maneuvers.

There is no assurance that the process described by this model fully accounts for all

adaptation phenomena; in fact, there are clear indications to the contrary (Guedry 1972) and the data obtained in this experiment suggested the possibility of additional processes. Nevertheless, the revised model represents a substantial improvement over the original torsion pendulum model in the prediction of responses to prolonged angular accelerations, and thus provides a rational basis for selection of adaptation measurements.

This report is based on work completed at the Naval Aerospace Medical Research Laboratory while the first and second authors were on active military duty in the U.S. Army and Navy respectively.

Opinions or conclusions contained in this report are those of the authors. They are not to be construed as necessarily reflecting the view or the endorsement of the Departments of the Army or Navy.

ZUSAMMENFASSUNG

Für Winkelbeschleunigungsreize von verhältnismässig kurzer Dauer werden die Reaktionen der Bogenorgane vom Torsionspendelmodell vorausgesagt. Bei längerer Dauer der Beschleunigungen treten jedoch gewisse Abweichungen auf. In einer ausserdings vorgeschlagenen, verbesserten Form des Torsionspendelmodells werden viele dieser Abweichungen mit einem kurzfristigen Anpassungsprozess erklärt. Zur Prüfung des umgearbeiteten Modells wurden dessen Vorhersagen mit den tatsächlich beobachteten Reaktionen bei Versuchspersonen verglichen, die einer Reihe von lange andauernden Beschleunigungen ausgesetzt worden waren. Das Modell beschrieb diese Ergebnisse ziemlich genau, obwohl unbedeutende Abweichungen bestehen blieben, was auf zusätzliche Anpassungsprozesse hinweist.

Eine neue Methode zur Messung des vom Modell beschriebenen Anpassungsprozesses wird dargestellt. Diese Methode beruht auf einer Einzelmessung, die verhältnismässig leicht entweder für Nystagmus oder für Empfindungsreaktionen durchgeführt werden kann.

REFERENCES

- Aschan, G. & Bergstedt, M. 1965 The genesis of secondary nystagmus induced by vestibular stimuli. *Acta Soc. Med. Upsal* 60, 113.
- Collins, W. E. & Guedry, F. E. 1962 Arousal effects and nystagmus during prolonged constant acceleration. *Acta Otolaryng (Stockh.)* 54, 349.
- Van Egmond, A. A. J., Groen, J. J. &

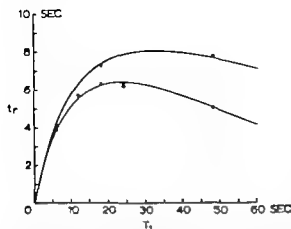


Fig 2 t values for nystagmus (●) and for sensation (○) to stimuli of various durations (6 subjects \times 6 trials). Also shown is a description by Eq 1 of nystagmus data with $\tau = 16$ sec and $\tau = 150$ sec, and a description of sensation data with $\tau = 16$ sec and $\tau = 50$ sec

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Thus, t depends upon the values of two parameters— τ and τ . When τ is known, τ for sensation can be determined.

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Eq 1 specifically predicts that, as T_1 increases, the t interval will grow longer until it reaches a maximum and then it will begin to grow shorter. Furthermore, if the subjective response does in fact adapt faster than nystagmus, the t for sensation should reach a maximum sooner than t for nystagmus and then decline more rapidly. The data, shown in Fig. 2, confirm all of these predictions. With τ set at 16 sec, an acceptable fit with the nystagmus data was obtained when τ for nystagmus was 150 sec, and a fit with subjective data was obtained when τ for sensation was 50 sec. Both values are somewhat similar to those employed by Young & Oman (1969) to describe data from other sources. The accuracy of the prediction thus offers additional support for the

THE INFLUENCE OF DRUGS ON CALORIC INDUCED NYSTAGMUS

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(Received April 15 1972)

Abstract. The influence of both the activating and the inhibitory power of voluntary fixation upon caloric nystagmus has been studied. Eight normal test subjects were given bithermal caloric irrigations to evaluate the influence of fixation and no fixation under various test conditions. The following effects were observed:

(1) Eye closure (i.e. with no fixation) has a facilitative power on the nystagmus in normal persons but inhibits or abolishes the induced nystagmus under the influence of reticular formation-blocking drugs.

(2) Eye opening (with fixation) diminishes or abolishes the nystagmus induced in normal persons, and increases or revives the nystagmus in persons given the reticular formation blocking drugs.

(3) It is concluded that, when the caloric test is performed, the resultant nystagmus should be examined under each of the two conditions of eye closure (no fixation) and eyes open (fixation). This may add useful diagnostic information to the otoneurological evaluation of patients. Electronystagmographic examination conducted only with the eyes closed may provide misleading information concerning vestibulo-ocular function.

The concept that the vestibulo-ocular reflex arc has more than one neuronal pathway has been accepted for a number of years (Lorente de N6 1933 1938 Spiegel & Price, 1939 Szentágothai, 1950 Szentágothai & Scháb 1956 Bender 1955 McCabe 1965). The medial longitudinal fasciculus has been accepted as the basic neuronal structure of the elementary three-neuron vestibulo-ocular mechanism. Its role, however, is apparent only in contrac-

tive responses (Lorente de N6, 1933 Szentágothai, 1950 Szentágothai & Scháb, 1956). The second pathway as was first shown by Lorente de N6 (1933, 1938) consists of inter-nuncial chains within the reticular formation.

The specific effect of certain drugs on the reticular formation has been reported by several authors (Moruzzi & Magoun, 1949 Bradley & Elkes, 1957 McCabe 1965 Akema et al. 1966). Its influence on the quick component of vestibular nystagmus has been demonstrated by Bender (1955) Bender et al. (1955) Anderson et al. (1958) Nathanson & Bergman (1958), Jongkees & Philipszoon (1960), Jatho (1965) McCabe (1965). There exist drugs which act directly on the reticular formation, either by a depressant action such as the barbiturates (Moruzzi & Magoun, 1949 Bradley & Elkes, 1957 Killam & Killam 1958) and alcohol, or by an excitant action, e.g. amphetamine (Bradley & Elkes, 1957). The barbiturates are believed by some authors (Moruzzi & Magoun, 1949 Bradley & Elkes, 1957) to block the conduction in the medial pathways at the brain stem level. Unconsciousness, sleep and the upper level of anesthesia block the quick component of nystagmus, the eyes move to the side of the slow component, and the induced nystagmus is never completed (McIntyre, 1939 Nathanson & Bergman, 1958 Blegvad, 1962).

In recent years, the ocular influence upon vestibular nystagmus has attracted many

searchers. Clinical experience of routine caloric testing of the vestibular organ has shown that, with fixation of gaze, there is a suppression of nystagmus, both with respect to the speed of the slow component and the duration (Aschan et al. 1956 Henriksson, 1956 Ma honey et al. 1957). There are cases in which nystagmus may be abolished completely by fixation. When the eyes are closed and fixation is abolished, the nystagmus will appear enhanced in all its features (Collins, 1962, 1968 Jongkees & Phillipszoon, 1964 Sokolovski, 1963 1966).

However the existence of a nystagmus which is primarily and possibly only seen on fixation and its disappearance under the conditions which diminish fixation has been known since Holmes' publication on acute cerebellar injuries in 1917 (Holmes, 1917 Rashbass & Russell, 1961 Naito et al. 1963 Dix & Hallpike 1966 Hart, 1967). Thus, the problem of the enhancing or abolishing power of ocular fixation on nystagmus appears to be still unsolved.

In an attempt to throw more light upon this phenomenon, the present investigation was carried out. Using normal subjects, we examined the influence of voluntary fixation upon induced vestibular nystagmus and the influence of drugs that are believed to act on the brain reticular formation.

METHOD

Subjects

Eight young subjects, who were all university graduates or students, were employed for this study. None had a history of ear trouble or vertigo. All were examined audiometrically to establish that their hearing was normal. All eight had otoscopically normal ear canals and tympanic membranes.

Examination of equilibrium and for both spontaneous and positional nystagmus were performed on each subject and found to be normal.

Apparatus

The thermal stimulus was provided by a thermostatically controlled water bath to provide temperatures of 44 C and 30 C at the end of the tube. The electronystagmograph recording apparatus was the Elema direct writing mingograph, M34. A time constant of 5 sec and a paper speed of 10 mm per sec were used. Surface electrodes were placed at each outer canthus of the subject's eyes to record the horizontal component of eye movements. A ground electrode was taped to the forehead. The whole unit was calibrated by asking the subject to follow two flashing lights 10 apart on the ceiling. The calibration was repeated after each irrigation. The sensitivity of the recording apparatus was adjusted to give a pen deflection of about 1 mm per 1 eye movement.

Procedure

The bithermal caloric test advocated by Fitzgerald & Hallpike (1942) was employed. The period of irrigation lasted 40 sec. The subjects were tested in a semi-dark room and, in addition, throughout all tests, a serial seven subtraction task was performed. Initially the induced nystagmus was recorded with the eyes closed (i.e. without fixation). After a period of 80 sec (from the onset of irrigation) the subject was instructed to open his eyes and look for a period of 20 sec at an illuminated point on the ceiling. The point was in the direct line of gaze and at a distance of 1.27 meters from the subject's eyes. After this 20 sec period, the subject was again instructed to close the eyes and the nystagmus was recorded until it disappeared. Following each caloric test optomotor nystagmus was recorded.

The eight normal subjects were submitted to caloric stimulation under five different test conditions. An interval of 7 days separated each test. The order of the tests was as follows: (1) Control test. (2) After administration of alcohol. 100 mg of gin was given orally 30

min prior to the test (3) After administration of barbiturate: 200 mg of sodium amylbarbitone was ingested 1 hour prior to the test. (4) After administration of amphetamine. 5 mg of *d*-amphetamine was administered orally 30 min prior to the examination (5) Final control test.

Analysis

Only the data on responses following irrigations of the right ear with water at 44°C were analysed. An analysis was made of each nystagmic reaction with eyes closed (i.e. without fixation) and with the eyes open (i.e. with fixation) during the culmination period of the caloric reaction. Two indices of response were used: (1) Stable's (1956) measure of maximum velocity and (2) frequency of the induced nystagmus. The velocity measure was calculated by taking the mean eye speed during a 10 sec period at the peak of the response (Stable 1956, 1958). The frequency was obtained by counting the number of beats over a 10 sec period at the culmination of the reaction (Torok, 1967 Hinchcliffe, 1968)

RESULT

The mean values were calculated for each of the two indices and under the different test conditions (Figs. 1 and 2)

In the control tests (drugs not administered) the velocity measure of the induced nystagmus with eyes closed (i.e. without fixation) was markedly increased in comparison with the nystagmus recorded with the eyes open (i.e. with fixation). The introduction of active visual fixation resulted in significant reduction in the velocity and frequency measures (Figs. 3 and 2).

Alcohol ingestion

The results of the test with alcohol (Fig. 4) showed very irregular caloric-induced nystagmus during eye-closure as compared with that with the eyes open (i.e. fixating) The mean velocity and frequency values, however

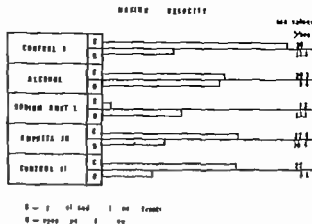


Fig. 1 Mean values of maximum velocity (expressed in degrees per second) of postcaloric nystagmus during eye closure (no fixation) and eye opening (fixation) for temperature 44°C, right ear, different test condition

are almost equal during eyes open and eyes closed (Figs. 1 and 2)

Barbiturate ingestion

Following ingestion of barbiturates, the nystagmus recorded with eyes closed failed to appear at all in many instances, or if present, was irregular and the beats weak (Figs. 5 and 6). With the eyes open and fixating, one can see well developed, regular nystagmus. However the velocity measure of the nystagmus

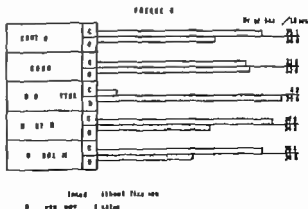


Fig. 2 Mean values of frequency (expressed in mean number of beats per 10 seconds) of postcaloric nystagmus during eye closure (no fixation) and eye opening (fixation) for temperature 44°C, right ear, different test condition.

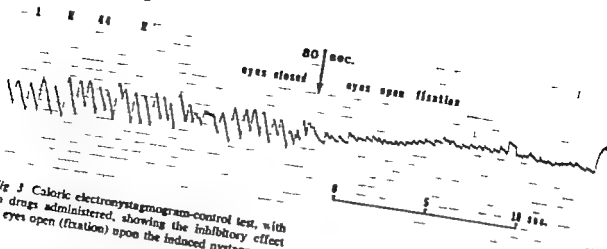


Fig 3 Caloric electronystagmogram-control test, with no drugs administered, showing the inhibitory effect of eyes open (fixation) upon the induced nystagmus.

during fixation was not essentially different from that of the induced nystagmus during fixation in subjects where barbiturates were not administered (Fig. 1) the frequency measure was much higher

Amphetamine ingestion

Here the nystagmus observed with the eyes closed is greater than that observed with the eyes open (Figs. 7 1 and 2) The effect of amphetamine was to improve the regularity of both the frequency and the amplitude of nystagmus during eye closure. With the eyes open no difference was observed in either the velocity or the frequency measures of the induced nystagmus as compared with the control tests.

The average values for the two measures of nystagmic response for the five conditions are shown graphically in Figs. 8 and 9. An

analysis of variance for the results obtained under the five test conditions showed that, for both velocity and frequency measures of the induced nystagmus responses where drugs had been given were significantly different from the control responses ($p > 0.001$).

DISCUSSION

These experiments have shown that, under the influence of sodium amylobarbitone, which is believed to block the reticular formation, caloric stimulation does not induce any consistent wave forms of nystagmus when examination was performed with eyes closed (i.e. no fixation). However with the eyes open and attempted fixation the appearance of vestibular nystagmus, which was not previously apparent was striking (Figs. 1 2, 5 6, 8

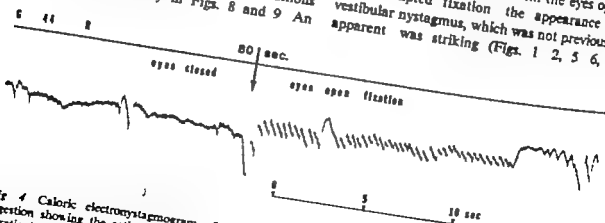
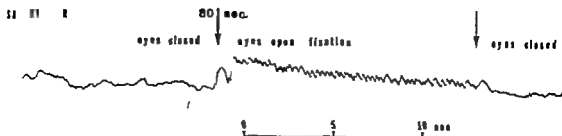
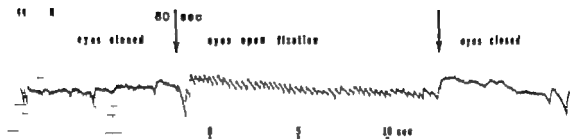


Fig 4 Caloric electronystagmogram after alcohol ingestion showing the activating effect of eyes open (fixation) upon the induced nystagmus.



5



6

5 and 6 Caloric electro-nystagmogram after Sbarate ingestion showing the activating effect of eyes open (fixation) upon the caloric nystagmus. The nystagmus is not present or very faint and dysrhythmic

when the eyes are closed and fixation abolished. The subject was performing a serial seven subtraction task throughout the test.

d 9). The activating influence of fixation seen also after alcohol ingestion (Fig. 4)

Thus, different results are observed in Tests 4 and 5 (Figs. 3 and 7). Eye closure brings activation of nystagmus in all its features. With the eyes open and fixating, an inhibitory effect can be seen upon the induced nystagmus.

What is therefore demonstrated here is a dynamic change in the form of the response. This effect may be termed "fixation activation"

of absent or diminished nystagmus, and the other "fixation inhibition" of previously present nystagmus.

The inhibitory influence of fixation on vestibular reactions is well recognized (Periman & Case 1939, Aschan et al. 1956, Henriksson 1956, Collins, 1968, Sokolovski, 1963, 1966 and many others). It has been shown that all features of the nystagmus are greatly reduced under the influence of fixation (Sokolovski 1963). Our results confirmed this. Weak nys-

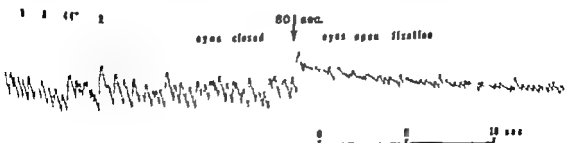


Fig. 7 Caloric electro-nystagmogram after amphetamine ingestion showing the inhibitory effect of eyes

open (fixation) upon the nystagmus. Notice ~~weak~~ nystagmus with eyes closed

MAXIMUM VELOCITY

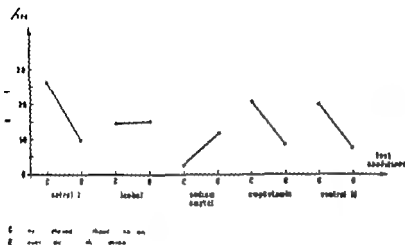


Fig 8 Group means, averaged across ears, of maximum velocity of postcaloric nystagmus expressed in degrees per second during eye closure (no fixation) and eye opening (fixation) conditions in the five test procedures.

tagmic responses were recorded in normal subjects when the fixation was introduced, whereas no-fixation and eye closure resulted in increased intensity of the caloric nystagmus.

If the nystagmus mechanism is dependent upon an interaction between vestibular nuclear neurons and reticular formation neurons, and if most of the vestibular impulses are conducted through the reticular formation (Gernandt, 1968) then the action of the specific drugs on the reticular formation should affect mainly the impulses funnelled through this pathway. The impulses funnelled through the medial longitudinal fasciculus will be unaffected. This means that only the discharges able to elicit the slow phase are not blocked (Lorente de No 1933, 1948 Szentágothai 1950 McCabe

1965). This was the condition in our experiments with sodium amytal where, during eye closure and no fixation very little or no induced nystagmus was encountered the eyes being held in tonic deviation in the direction of the slow component. Yet clear nystagmus, with its slow and quick component appeared during eye opening and fixation. The intensity of this nystagmus, however was never equal to the intensity of the nystagmus with eyes closed in the same subjects, when not under the influence of amytal.

Based on our data obtained from the tests carried out on the 8 normal subjects, it is postulated that, on eye opening (fixation), the reversal (quick phase) of the slow component

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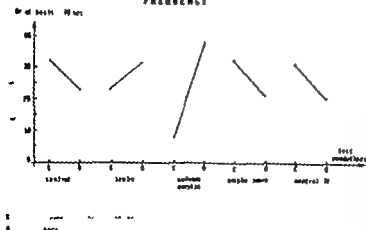


Fig 9 Group means, averaged across ears, of frequency of postcaloric nystagmus expressed in number of beats per 10 seconds during eye closure and eye opening conditions in the five test procedures.

the caloric nystagmus is acting along with the stimulation of attempted fixation. Consequently summation of the two separate strong stimuli opposing the slow component will result in an inhibition of the nystagmus. Thus nystagmus, if manifested, may be of more regular frequency but of smaller amplitude and/or intensity.

In our experiments with sodium amytobarbital the reticular formation, and therefore the quick component of the induced nystagmus is believed to be blocked. However the vertical longitudinal fasciculus is unaffected and able to conduct the slow component. Therefore the eyes would deviate in the direction of the slow phase and remain there. On opening and attempted fixation the rapid movement toward the straight ahead position swings the eyes back to the mid-line, but the deviating force of the slow component is still acting and so the nystagmic rhythm is established until the cupula can return to its zero position or upon eye closure when the force of attempted fixation ceases to act, the nystagmus would disappear. In a clinical routine this phenomenon of increased nystagmus with eyes open and fixation is observed fairly frequently and has been reported in connection with posterior fossa lesions by Holmes (1917), and later by others (Moberg et al., 1962; Naito et al., 1963; Dix & Hallpike, 1966; Edoux & Demancez, 1967; Hart, 1967; Coats, 1970).

If then, eye closure with no fixation is taken as a standard state, eye opening and fixation is found to have an inhibitory effect upon caloric induced nystagmus in normal persons and an activating effect upon this nystagmus under the influence of the reticular formation blocking drugs. We believe that it is the attempted fixation which brings the nystagmic rhythm back.

It is our impression that the above electro-nystagmographic pattern might be of great diagnostic significance in brain stem lesions. We might propose that the inhibition of the nystagmus during eyes open fixation be termed

"Fixation Inhibition" (F.I.) and the enhancement, or the recurrence, of the nystagmus previously not present, be called "Fixation Activation" (F.A.) The latter would be characteristic of central vestibular lesion involving the brain stem—reticular formation, providing drug influence could be excluded, whereas the former pattern is indicative of normal vestibular function or of peripheral involvement if there is a vestibular disorder.

ACKNOWLEDGMENTS

I wish to acknowledge my gratitude to Dr J. E. Bondley and Dr G. T. Nager for their interest and help in this work, and to Dr R. Hinchcliffe for his kind assistance and criticism.

ZUSAMMENFASSUNG

Untersuchungen über die gesteigerte oder hemmende Einwirkung optischer Fixation auf den kalorischen Nystagmus wurden durchgeführt. Acht normale Versuchspersonen erhielten intravenöse, kalorische Spülungen, um den Einfluss der Fixations- oder Nichtfixationsmethode unter verschiedenen Versuchsbedingungen zu bestimmen.

Folgende Beobachtungen wurden gemacht:

(1) Schliessen der Augen ohne Fixation hat gesteigerte Wirkung auf den Nystagmus von Normalpersonen und hemmt oder unterdrückt Nystagmus unter dem Einfluss von Drogen mit Hemmwirkung auf die retikuläre Formation.

(2) Öffnen der Augen und Fixation verringern oder unterdrücken den Nystagmus bei normalen Versuchspersonen und steigern oder reaktivieren den Nystagmus bei Personen unter Einfluss von Drogen, die die retikuläre Formation blockieren.

(3) Man kann daraus schliessen, dass bei Durchführung kalorischer Tests der Nystagmus unter beiden Bedingungen — Augen geschlossen (keine Fixation) und Augen geöffnet (Fixation) — untersucht werden muss. Dadurch bekommt man wertvolle diagnostische Hinweise für die otoneurologische Untersuchung der Patienten.

Wird die elektronystagmographische Untersuchung dagegen nur bei geschlossenen Augen ausgeführt, können daraus leicht falsche Schlüsse hinsichtlich der vestibulo-okulomotorischen Funktion gezogen werden.

REFERENCES

- Aleksis, G., Pernis, L., Rosenthal, G., Rowl, G. F. & Zatz, J. 1966 Functional inactivation of the brain stem related to the level of consciousness. *J. Neurosurg.* 24: 629.

- Anderson, P J Diamond, S. P Bergman, P S. & Nathanson, M 1958 Electrooculographic investigation of the caloric response *Neurology* 8 741
- Ahn, G Bergstedt, M & Stahl, J 1956 Nystagmograph recording of nystagmus in clinical neuro-otological examinations. *Acta Otolaryng* (Stoch.) Suppl. 179 1
- Bender M B. & Bergman, P S. 1955 Neuro-ophthalmology (Nystagmus). *Prog Neurol Psychiat* 10 01
- Bender M B 1955 The eye-centering system. *Arch Neurol Psychiat* 73 685
- Bjergvad, B 1966. Caloric vestibular reaction in unconscious patients. *Arch Otolaryng* (Chic.) 75 506
- Bradley P B & Ellis, J 1957 The effects of some drugs on the electrical activity of the brain. *Briefs* 80 77
- Coats, A. 1970. Central electrical nystagmographic abnormalities. *Arch Otolaryng* 73 685
- Collins, W E. 1966. Control of nystagmus by manipulation of the distance. *Ann Otol* 71 187
- 1968 Special effects of nystagmus. *Aerospace Med* 39 5
- Dit, M. R. & Hallpike (the clinical features of spontaneous nystagmus lateral acoustic neuroma) (Stockh.) 61 1
- Fitzgerald, G & Hallpike (human vestibular function) (Stockh.) 61 1
- Germant, B. E 1968. The myotatic and ascending nystagmus. *Neurol* 20 170
- Hart, C W 1967. Ocular nystagmus. *Laryngoscope* 77 101
- Henriksson, N G 1956. Speed of nystagmus and duration in caloric nystagmus. *Acta Otolaryng* (Stockh.) Suppl. 179 3
- Hill, R 1968. Nystagmus test in an index of caloric response. *Acta Otolaryng* (Stockh.) 63 311
- Holmes, G 1917. The symptoms of acute cerebellar injuries due to gunshot wounds. *Brain* 40 461
- Jahko, A von 1965. Die Wirkung der Alkoholintoxikation auf den Vestibularapparat mit besonderer Berücksichtigung der Störungen der vestibulären Otolithfunktion. *Z Laryng Rhinol Otol* 44 1
- Jongkees, L B & Philipsson, A. J 1964. Electro-nystagmography. *Acta Otolaryng* (Stockh.) Suppl. 189 7
- 1960. Some nystagmographical methods for the investigation of the effect of drugs upon the labyrinth. *Acta Physiol Pharmacol Neurol* 9 40
- Kilian, A. F & Kilian, E A. 1958. *Retikular formation of the brain* (ed H H Jasper). Little, Brown & Co Boston
- Ledoux, A. & Demaree, J P 1967. Nystagmographie les yeux ouverts ou fermés au cours de l'épreuve calorique. *Acta Otolaryng* 8 17 31
- Lorente de No, R. 1933. Vestibulo-ocular reflex arc. *Arch Neurol Psychiat* 30 245
- 1938. Analysis of the activity of the chains of internuncial neurons. *J Neurophysiol* 1 207
- Maloney, J L., Harlan, W L. & Bickford, R G 1957. Visual and other factors influencing caloric nystagmus in normal subjects. *Arch Otolaryng* (Chic.) 66 46
- McCabe B F 1965. The quick component of nystagmus. *Laryngoscope* 75 1619
- McIntyre A. K. 1959. The quick component of nystagmus. *J Physiol* 97 8
- Moberg, A., Preber, L., Silfverklöf, B. & Valbo, A. 1966. Imbalance nystagmus and diplopia in Wallenberg's syndrome. *Acta Otolaryng* (Stockh.) 55 69
- Moruzzi, G & Magoun, H. W 1949. Brain stem reticular formation and activation of the EEG. *Electroencephalogr Clin Neurophysiol* 1 455
- Naito T., Tatum, T., Matsunaga, T. & Matsunaga, T 1963. The effect of eye-closure upon nystagmus. *Acta Otolaryng* (Stockh.) Suppl. 179 72
- Nathanson, M. & Bergman, S. 1958. Newer methods of evaluation of patients with altered states of consciousness. *Med Clin N Amer* May p 701
- Perkins, H B. & Case T J 1959. Nystagmus: Some observations based on an electrical method for recording eye movements. *Laryngoscope* 69 17
- Rashbass, C. & Russell, G F M. 1961. Action of barbiturate drug (Amyobarbitone Sodium) on the vestibulo-ocular reflex. *Brain* 84 379
- Solomon, A. 1963. Factors influencing nystagmus due to rotation in normal patients. *J Laryng* 77 185
- 1966. The influence of mental activity and visual fixation upon caloric induced nystagmus in normal subjects. *Acta Otolaryng* (Stockh.) 61 709
- Spirogl, E. A. & Price J B 1939. Origin of the quick component of labyrinthine nystagmus. *Arch Otolaryng* (Chic.) 30 576
- Stahl, J 1958. Electronystagmography in the caloric and rotatory test. *Acta Otolaryng* (Stockh.) Suppl. 137 8
- 1956. Electronystagmography in the caloric test. *Acta Soc Med Upsal* 61 307
- Szentágothai J 1950. The elementary vestibulo-ocular reflex arc. *J Neurophysiol* 13 395
- Szentágothai, J & Schab, R. 1956. A midbrain inhibitory mechanism of oculomotor activity. *Acta Physiol* 9 89
- Torok, N 1967. How vestibular test results can be utilized. *Trans Amer Acad Ophthalm Otolaryng* May-June p 416-470.
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INTERACTION BETWEEN THE UTRICLES AND THE HORIZONTAL SEMICIRCULAR CANALS

III. Sectioning of the Horizontal Ampullar Nerve on One Side and of the Utricular Nerve on the Other Followed by Tilting around the Longitudinal Axis

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(Received October 30, 1973)

In 70 cats selective sectioning of the horizontal ampullar nerve was performed on one side, of the utricular nerve on the other. Sectioning the ampullar nerve caused nystagmus in the contralateral direction. This nystagmus either decreased or appeared completely as a result of the subsequent sectioning of the utricular nerve. Tilting toward the side where the ampullar nerve had been cut resulted in a decrease in the previously diminished nystagmus, and its reappearance, if it had previously been inhibited. Tilting in the other direction caused no reaction in those cases where inhibition had occurred and total inhibition in those cases where nystagmus had been decreased. Finally if both side nerves were cut there was no alteration in nystagmus during tilting in either direction.

Semicircular canals and the otolith organs are considered to perform different functions. They are moreover also stimulated differently: the former by angular, the latter by linear acceleration. However they do not operate independently of each other but seem to be mutually coupled in their function (Maxwell 1933, McNally 1955). In a previous paper the present authors (Fluor & Slegborn, 1973) showed that in cats whose horizontal ampullar nerve and utricular nerve had been subjected to unilateral selective sectioning tilting the animals around their longitudinal axis caused a change in the nystagmus, induced by the rotation. Tilting toward the operated side resulted in increased nystagmus frequency whereas tilting in the opposite direction caused

inhibition. Hence, it was concluded that the intact utricle facilitates nystagmus brought about by the ipsilateral horizontal semicircular canal. This has given rise to a new problem which will be dealt with in this paper.

What effect has the utricle on nystagmus induced by the contralateral horizontal semicircular canal?

MATERIAL AND METHOD

Twenty adult cats were used for the experiments. The operational- stimulative and recording techniques are approximately the same as those described in our previous article (Fluor & Slegborn 1973). For sectioning of the utricular nerve not only was a fenestration performed but the bone was removed right down to the oval window so that the horizontal ampullar nerve was clearly visualized. The caudal parts of the oval window were also drilled off. Thereafter the utricular nerve was approached by carefully drawing aside the horizontal ampullar nerve. It is also important to limit the upper border of the utricular nerve against the anterior ampullar nerve. The nerve is dissected free with fine needles and is close to the entrance into its bony canal. Great care must be taken not to pull it so that the membranous wall of

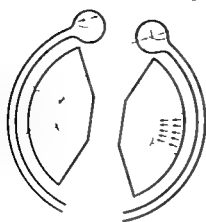


Fig. 1 Schematic diagram of the utricle and horizontal semicircular canals in resting position after selective sectioning of the right horizontal ampullar nerve and of the left utricular nerve. The arrows indicate the orientation of the hair cells, i.e., the direction in which they increase their discharge frequency.

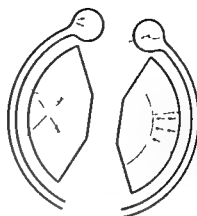


Fig. 2 Schematic diagram of the utricle and horizontal semicircular canals after selective sectioning of the right horizontal ampullar nerve and of the left utricular nerve, when the animal is tilted to the right. Arrow indication same as in Fig. 1. The number of arrows symbolizes discharge frequency.

becomes dislocated, thus damaging the entrance of the horizontal and anterior ampullae. The experiments were sometimes finished by cutting the nerve of the remaining utricle.

RESULT

Selective sectioning of the horizontal ampullar nerve resulted in a destruction nystagmus toward the contralateral side when the animals

had awoken to a level of superficial anesthesia. In six cats the subsequent selective sectioning of the contralateral utricular nerve (Fig. 1) gave rise to a considerable decrease in nystagmus frequency from about 15-25 beats/10 sec down to 4-10 beats/10 sec. In 14 animals, nystagmus was totally inhibited. In those subjects whose nystagmus frequency decreased

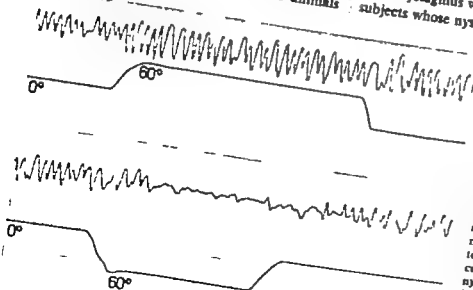
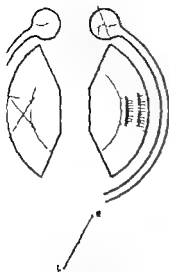


Fig. 3 Consecutive curves from a cat after selective sectioning of the right horizontal ampullar nerve and of the left utricular nerve followed by tilting to the right. The upper curve shows electro-nystagmography and the lower curve the tilting of the table. Tilting to the right upwards, to the left downwards.



4 Schematic diagram of the utricles and horizontal semicircular canals after selective sectioning of the right horizontal ampullar nerve and of the utricular nerve, followed by tilting to the left. The symbols are the same as in Fig. 1. The number of symbols indicates discharge frequency.

As a result of the operation, tilting toward the side where the ampullar nerve had been cut (Fig. 2) increased the nystagmus frequency by 2 beats/10 sec (Fig. 3). In two cases, tilting toward the other side (Fig. 4) produced a decrease in frequency of about 50% whereas in our cases there was total inhibition (Fig. 3). In those cats in which the operation caused total inhibition of nystagmus it reappeared (Fig. 5) during tilting toward the side where the ampullar nerve had been cut, whereas on tilting toward the other side, no nystagmus was observed.

Finally in six cases the nerve to the remaining utricle was cut. In those cats which

had previously had a slow nystagmus a decrease in frequency occurred when there had been total inhibition of nystagmus recurred with slow frequency. In some of these animals did not influence the direction of the nystagmus.

DISCUSSION

This operation creates a situation in which the activity of the horizontal semicircular canal is missing on one side and that of the utricle is missing on the other. It has shown that an increase in the activity of the "horizontal areas" of the utricle is brought about by tilting. The nystagmus induced by the horizontal semicircular canal is put from the utricle as a result of the nystagmus. These results show a crossed inhibitory interaction between the lateral utricle and the horizontal semicircular canal. This was obtained during sectioning of the horizontal semicircular canal (Fig. 1973). That the lateral utricle is also involved in the sectioning of the horizontal semicircular canal is shown by the fact that it released a nystagmus during further tilting.

Bei 20 Katzen wurde das N. ampullar N. utricular N. ampullar nach der Verminde- Abgeschalt.



Kippung nach der Seite wo der N. ampullaris abgeschnitten war resultierte in einer Steigerung der zu- oder herabgesetzten Geschwindigkeit des Nystagmus oder in einem Wiederauftreten bei vorherigem Schwund. Eine Kippung nach der anderen Seite hin erbrachte keine Veränderung im Füllen mit Nystagmuschwund, aber eine totale Inhibition in den Fällen mit herabgesetztem Nystagmus. Wurden schließlich beide N. utriculares abgeschnitten, erhielt man keine Veränderung der Nystagmus durch Kippung.

REFERENCES

- Fluor E. & Siegborn, J. 1973 Interaction between the utricles and the horizontal semicircular canals. I. Unilateral selective sectioning of the horizontal ampullar nerve followed by tilting around the longitudinal axis. *Acta Otolaryng* (Stockh.) 75 17
- 1973 Interaction between the utricles and the horizontal semicircular canals. II. Unilateral selective sectioning of the horizontal ampullar nerve and the utricular nerve followed by tilting around the longitudinal axis. *Acta Otolaryng* (Stockh.) 75 393
- Maxwell, S. S. 1923 *Labyrinth and equilibrium*. Lippincott, Philadelphia and London.
- McNally W. J. 1955 Some facts and fancies about the utricle. *Ann Otol* 64 355
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COCHLEAR MICROPHONICS RECORDED FROM THE EAR CANAL IN MAN

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By using average technique, cochlear microphonics were recorded from an electrode in the ear in response to 1 kHz-tone bursts, delivered to a metal, magnetically shielded TDH 39 ear in a closed acoustic system. No measurable difference could be recognized, either in normal ears or in deaf ears. The amplitude function is described 65 dB to 120 dB p.e. SPL and appears to be similar to the established pattern in animals. The phase of the cochlear microphonic was compared to the phase of the stimuli in the ear canal, and the influence of the stapedius muscle contraction on the amplitude and phase of the cochlear microphonic is described. Besides the cochlear microphonics the action potentials, a summing potential in response to 4 kHz-tone bursts was recorded. Cochlear microphonics in a few pathological ears are outlined. A latency of 14 msec a multiphasic response was often present. The results are dis-

non-surgical recording of cochlear microphonics in humans by an averaging technique. The present paper will report findings at 1 kHz only where the CM are most readily recorded.

METHOD

The recording techniques have been described earlier (Salomon & Elberling 1971, Elberling & Salomon, 1971). Differences from the previous reports were as follows: A 0.25 mm Teflon-insulated silver wire was placed with the chlorided tip in the skin of the ear canal next to the ear drum. A differential amplifier with 90 dB voltage amplification and a frequency response from 5-5 000 Hz was used. The input impedance was 1 G Ω and the common mode rejection, while using our electrodes, was greater than 120 dB. The equivalent input noise was 1.5 μ V p.p.

1 000 Hz tone bursts were delivered via a TDH 39 earphone shielded in a cylindrical μ -metal case of 0.5 mm thickness. In the top and the bottom of the case two extra 1 mm μ -metal discs were inserted, separated from the case by copperfoil. The cable near the earphone was also wrapped in μ -metal. The stimulus was a tone burst (normally 5 msec), which was measured using a 3-chamber artificial ear (B & K 4153) giving both the amplitude in dB p.e. SPL and the phase (Fig. 1). We have previously shown the approximate

cochlear microphonics (CM) were first described in animals (Wever & Bray 1930) and have been recorded in man by surgical methods from the round window (Perlman & Ruben et al., 1959) and also by non-surgical methods (Yoshie & Yamaura, 1969). Excellent literature reviews are found in their papers.

CM are related to the physiological integrity of the cochlear partition structures and can be used as an indicator of the degree of hair cell damage (Simmons & Beatty 1962). CM can also be used to analyse middle ear transmission (Møller 1963). This paper concerns

This work was supported by The Danish Medical Research Council.

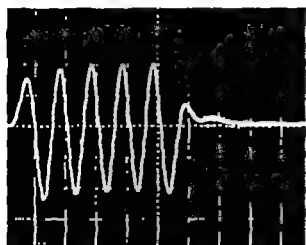


Fig. 1 The 1 kHz, 5 msec tone burst stimuli, measured in a 3-chamber artificial ear (Brüel & Kjær type 4153). Time scale: 1 msec/div

tween measurements of transient sounds made in this way and with a corrected sound probe (Salomon & Elberling, 1971)

RESULT

1. Evaluation of the μ -metal shielded ear phone

In our previous paper (Salomon & Elberling, 1971) we used an iron-shielded TDH 39 ear phone which produced an artifact of 500–1 000 nV p.p. at 1 000 Hz and 120 dB p.e. SPL on deaf ears. This artifact, having a power function of 1/3 showed a decrease with increasing frequency. Using the new magnetically shielded TDH 39 no artifacts could be detected in deaf ears. On testing the magnetic radiation from the μ -metal case a decrease of 26 dB at 1 000 Hz was found compared with the iron-shielded earphone. The absolute value of the magnetic field measured directly in front of the shielded earphone was 9×10^{-3} amp/meter at 1 000 Hz and 120 dB SPL and the shielding reduced the magnetic field by 45 dB compared with the bare TDH 39 earphone.

2. Measurement of CM in normal ears

In normal adults the CM could be recorded from 65 dB to 120 dB p.e. SPL.

At 85 dB the CM reached measurable values

of about 50 nV p.p. which was equal to the residual background noise after 2 000 averages. Steady tones are perceived 12 to 18 dB above the sensation of the corresponding 5 msec tone bursts at 95 and 0 dB SPL respectively (Pedersen & Elberling, 1972). The upper limit of the range studied thus corresponded to less than 110 dB SL.

Fig. 2 shows an example from a normal adult. From 65 dB to 90 dB p.e. SPL the amplitude function is almost linear but at 95 dB p.e. SPL the curve levels out. The CM reproduce the stimulating waveform at all intensities without appreciable distortion.

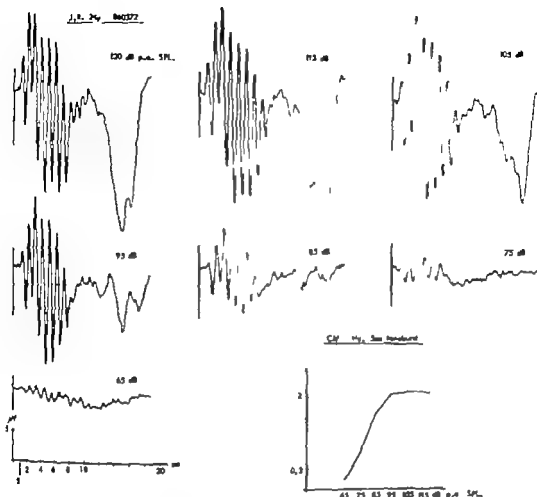
Fig. 3 also shows CM from a healthy person, but in this case the amplitude has a pronounced decrease at high intensity where a severe distortion of the CM is seen.

Fig. 4 shows the individual normalized amplitude functions from 5 normal persons, together with the mean curve. The mean amplitude curve shows a maximum at 105 dB p.e. SPL, a power function of 0.7 between 65 and 100 dB p.e. SPL and a decrease in amplitude above 105 dB p.e. SPL.

At a latency of 1.2–2.0 msec the CM are distorted by the action potential from the inner ear and at a latency of approximately 14 msec a multi-phasic potential appears. This potential (see Figs. 2, 3, 7) having a threshold of more than 80 dB p.e. SPL, shows decreased latency and increased amplitude with increasing intensity.

Fig. 5 demonstrates the phase-shift between the stimulating waveform and the CM in one recording. The average phase-shift in six different persons was 110° ranging from 90° to 180°.

In 1959 Galambos showed that middle-ear muscle contraction elicited by contralateral sound-stimulation reduced the CM in cats, and Fig. 6 shows that this is also the case in man. In a normal person the CM were recorded using 20 msec tone bursts, and the amplitude function was determined at 4 intensities. The reduced CM at 115 and 90 dB p.e. SPL caused by contralateral stimulation with 105 dB SPL.



2. CM recordings and the corresponding amplitude function in response to 1 kHz, 5 msec stimuli in a normal person.

to noise were recorded, and the new amplitude function is outlined. This amplitude reduction indicates a 9 dB reduction in the cochlear input.

Fig. 5 demonstrates that, besides the amplitude reduction, a phase-shift is produced by unilateral sound stimulation. In this recording the shift amounts to 35°.

Summating potentials were small at 1000 Hz, but increased with increasing frequency. Fig. 7 shows an example of this electro-negative potential recorded at 4000 Hz.

Measurement of CM in pathological ears

In pathological ears CM could also be recorded. Recordings from a postoperative oto-

sclerous (high tone loss) and a Mondini-like defect (basal turn intact low tone loss) are shown in Fig. 8 A and B, as examples of cochlear localized lesions in either the high or the lower frequency range.

Fig. 8 C is from a patient with a flat 60 dB cochlear hearing loss (Menière).

In 5 severely deaf ears only small CM amplitudes could be recorded. An example is shown in Fig. 8 D. The amplitude functions having no maxima showed a great spread between the slopes.

DISCUSSION

The fact that the CM approximately follows the stimulating waveform implies

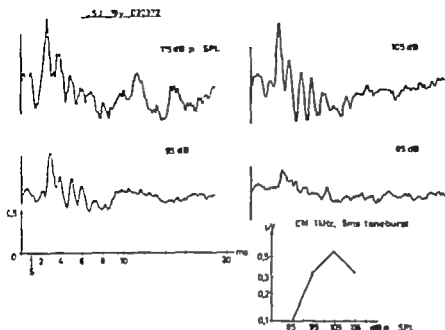


Fig 3 CM recordings and the corresponding amplitude function in response to 1 kHz, 5 msec stimuli from a normal person.

difficulties in differentiating the CM from artifacts. Electric and magnetic artifacts are approximately inversely proportional to the square of the distance between electrodes and sound transducer and Yoshie & Yamaura (1969) excluded artifacts using a distance of 30 cm. But in our instrumentation, proper shielding, high common mode rejection and low electrode impedance abolished any measur-

able artifacts, even when using a closed acoustic system on deaf ears. The electrode placement, avoiding contact with any acoustically active structures, did not give rise to mechanical artifacts, so that even when the vib-

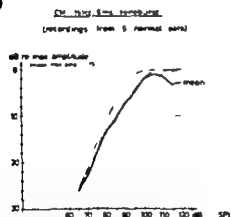


Fig 4 Double logarithmic plots of the CM amplitude functions from 5 normal persons, using 1 kHz, 5 msec stimuli (---). The individual curves have been normalized for comparison and the mean curve (—) is indicated.

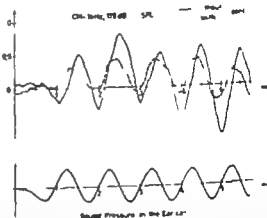


Fig 5 Phase relations of the CM waveforms. Top: CM recordings from a normal person, using 1 kHz, 70 msec stimuli at 115 dB p.e. SPL. Without (—) and with (---) constant contralateral 108 dB SPL white noise stimulation. Bottom: Waveform and phase of the stimuli in the ear canal, measured in a 3-chamber artificial ear (Brüel & Kjær type 4153). A phase difference between the stimulus and the "normal" CM (—) of 96 (about 265 μ sec) and a phase difference between the two CM curves of 35 (about 100 μ sec) is calculated.

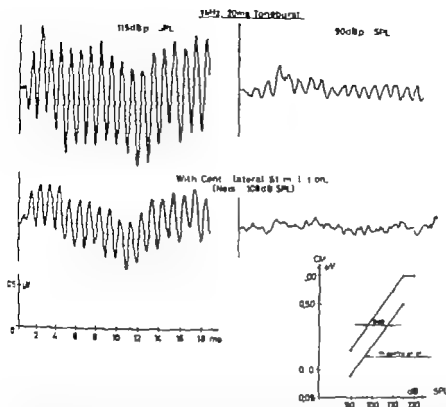


Fig. 6. Using 1 kHz, 20 msec stimuli at 4 intensities, in a normal person, the fig. shows (top) the recorded CM at 115 dB and 90 dB p.e. SPL. The calculated amplitude function (at 4 intensities) is shown (upper curve). Centre CM recordings are shown at the same

intensities, but with additional constant contralateral 108 dB SPL white noise stimulation (stapedius muscle contraction) giving a new amplitude function as shown (lower curve). The intensity shift between the two amplitude functions is about 9 dB as indicated.

tion properties of the electrode were increased in one experiment by coiling the wire in the ear canal, no artifacts were seen in deaf ears.

The use of narrow band filters (Gavilan & Sengco, 1964; Ruben, 1967) and the use of cut-off frequencies closer than 2 octaves from the measured CM (Yoshie & Yamaura, 1969) have been avoided to allow proper measurements of the phase of the CM.

The nature of our recordings being genuine CM seems to be established by the power-function of the measured CM corresponding to the well-known non-linear amplitude function (Wever & Bray 1938) the phase angle between the stimulating sound in the ear canal and the CM corresponding to the values reported in animals (Møller 1963) and

the fact that CM pick-up is changed in amplitude and phase by contralateral sound stimulation (stapedius muscle contraction).

By using both the so-called differential electrode technique (review Dallos, 1972) and a gross electrode near the round window in animals (Simmons & Beatty 1962) and in man (Yoshie & Yamaura, 1969) the recorded CM only reflect the cochlear generators within few mm of the electrode placement. An electrode in the ear canal with a more uniform distance to all parts of the cochlea, probably gives a more general picture of the overall cochlear functional state. Correspondingly the CM was close to normal values both in a patient with a pure cochlear high tone loss and in a patient with a low and middle frequency hearing-loss (Fig. 8, curve

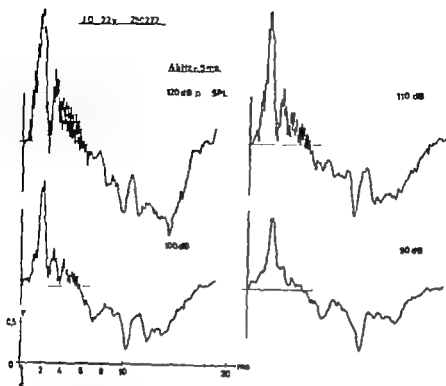


Fig 7 A negative summing potential recorded in response to 4 kHz, 5 msec tone bursts at 4 intervals.

whereas the CM were greatly reduced in patients where all frequencies were affected (Fig. 8, curves C and D). This seems to indicate that by using 1 000 Hz stimulation, the re-

corded CM provide information from any remaining normal part of the cochlea. This, combined with the fact that the CM are a logarithmic function with no threshold value, makes the clinical application difficult to assess.

In ears with severe and widely distributed cochlear lesions the recorded CM were small. In these cases the CM recordings can be used clinically.

The late multiphasic potential appearing after the CM (Figs. 2, 3, 7) is probably myogenic in origin. It has a latency of 10–15 msec, and it appears in shape similar to the responses reported by Kiang et al. (1963) and Borsanyi (1964). Like the sound-elicited stapedius muscle reflex, the threshold of the response is greater than 60 dB SL.

ZUSAMMENFASSUNG

Mit Hilfe der average technique wurden Cochlear mikrophonien — als Reaktion auf 1 kHz-Tonimpulse — registriert. CM wurden von einer Elektrode im inneren Gehörgang abgeleitet. Die Reaktion wurde von einem p-Metall auf magnetisch abgeschirmte TDH IV

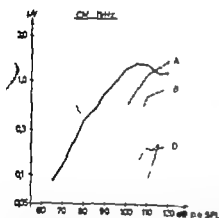


Fig 8 Normal and pathological CM-amplitude functions recorded at 1 kHz, 5 msec tone bursts. — mean, and dashed area, range covered by the recordings from 5 normal persons. results from pathological ears: A: postoperative otosclerosis with a cochlear high tone loss. B: Mondini-like defect (basal turn intact) with normal hearing in the high frequency range (6–10 kHz) and severe cochlear loss in the low frequency range. C: Menière-disease with a flat cochlear hearing loss of 60 dB HL. D: a cochlear hearing loss exceeding 100 dB HL at all frequencies.

dephon in einem geschlossenen akustischen System befindet. Sowohl bei Schwerhörigen als bei Normalhörenden konnten keine messbaren Artefakte erkannt werden. Die Funktion der Amplitude von 65 dB bis 70 dB p.a. SPL wurde beschrieben als ähnlich der eigenen, die bei Tierversuchen erzielt wurde. Die Lage der Cochlearmikrophonien wurde mit der Stimulationsphase im Gehörgang verglichen, und der Einfluss der Stapediusmuskulaturkontraktion auf die Lage und Amplitude der Cochlearmikrophonien wurde kurz beschrieben. Außer der Cochlearmikrophonien und der Aktiopotentialreize wurde auch eine Aktionspotentialreize als Reaktion auf 4 kHz-Tonpulse registriert. Die Cochlearmikrophonien bei leicht Schwerhörigen wurden erwähnt. Bei einer Latenz von ungefähr 14 msec ist oft eine mehrphasische exponente vorhanden gewesen. Die Ergebnisse werden behandelt.

REFERENCES

- Jonsson, S. J. 1964 Some aspects of auditory evoked potentials in man. *Ann Otol* 73 61.
- Liberman, P. 1972. Cochlear Potentials. A Status Report. *Audiology* 11 29.
- Elberling, C. & Salomon, G. 1971 Electrical potentials from the inner ear in man, in response to transient sounds generated in a closed acoustic system. *Rev Laryng (Bord)*, Suppl. 691.
- Strombos, R. & Rupert, A. 1959 Action of the middle ear muscles in normal cats. *J Acoust Soc Amer* 31 3.
- Wiken, C. & Svanborg, J. 1964 Microphonic potentials picked up from the human tympanic membrane. *Ann Otol* 73 1.
- Gang, N. Y., Christ, A. H., French, M. A. & Edwards, A. G. 1963 Postauricular electric response to acoustic stimuli in humans. *Quart Progr Rep Res Lab Electronics M.I.T* 68 218.
- Müller, Aa. R. 1963 Transfer function of the middle ear. *J Acoust Soc Amer* 35 10.
- Pedersen, C. B. & Elberling, C. 1972. Temporal integration of acoustic energy in normal hearing persons. *Acta Otolaryng* in press.
- Perlman, H. B. & Case, T. J. 1941 Electric phenomena of the cochlea in man. *Arch Otolaryng (Chic.)* 34 710.
- Ruben, R. J. 1967 Cochlear potentials as a diagnostic test in deafness. In *Sensorineural hearing processes and disorders* (ed. A. Bruce Graham), Chap. 25 Little, Brown & Co. Boston.
- Ruben, R. J., Knickerbocker, G. G., Sekula, J., Nager, G. T. & Bordley, J. E. 1959 Cochlear microphonics in man. *Laryngoscope* 69 665.
- Salomon, G. & Elberling, C. 1971 Cochlear nerve potentials recorded from the ear canal in man. *Acta Otolaryng (Stockh.)* 71 4.
- Simmons, F. B. & Beatty, D. L. 1962. The significance of round-window-recorded cochlear potentials in hearing. *Ann Otol* 71 3.
- Wever, E. G. & Bray, C. W. 1930 Auditory nerve impulses. *Science* 71 215.
- 1938. Distortion in the ear as shown by the electrical responses of the cochlea. *J Acoust Soc Amer* 9 227.
- Yoshida, N. & Yamasaki, K. 1963 Cochlear microphonic responses to pure tones in man recorded by a non-surgical method. *Acta Otolaryng (Stockh.)* Suppl. 252 37.

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VOCAL CORD FIXATION IN LARYNGEAL CARCINOMA

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(Received May 3 1972)

Abstract Fixation of the vocal cord in glottic carcinoma is a poor prognostic sign. In a follow-up study of 134 patients with T₂N₀M₀ glottic carcinomas, only 55% of those with impaired mobility of the vocal cord survived 5 years, compared with 70% of those with normal mobility. In a separate study of 48 serially sectioned laryngectomy specimens, with a fixed vocal cord before surgery the main cause of vocal cord fixation was invasion of the thyro-arytenoid muscle. The more extensive carcinomas invaded other laryngeal structures, including the laryngeal framework. There was a very poor correlation between the pre-operative clinical assessment and the pathological findings. Over 90% of these cases were clinically under-assessed, chiefly because of spread of the tumour outside the larynx. We have concluded that until better methods of clinical assessment are developed fixation of the vocal cord in glottic carcinoma is a contraindication to conservation surgery.

Fixation of the vocal cord has been regarded as a poor prognostic sign. Helse & Baylis (1966) found that 70% of the patients with T₂N₀M₀ glottic carcinomas with normal mobility of the vocal cord survived 5 years, compared with only 57% of those with fixation. Shaw (1965) Mårtensson et al. (1967) and Vermund (1970) presented similar figures. In order to test the validity of this concept, we studied the five year survival of 134 patients with T₂N₀M₀ glottic carcinomas treated in

Toronto. Our study confirms the earlier reported results.

In addition we studied 28 serially sectioned laryngectomy specimens from patients who had fixation of the vocal cord at the time of operation. These cases were selected from a larger series of laryngectomy specimens that have been processed in the same manner. The purpose of this study was to review the causes of vocal cord fixation. We found, as did Kirchner (1969) in a similar series, that the main cause of vocal cord fixation was invasion of the thyro-arytenoid muscle by tumour. We also wanted to compare the clinical pre-operative assessment with the histopathological findings in these cases. Many of the tumours were clinically under-assessed because of inability to assess spread outside the laryngeal framework. Whereas there are differing opinions as to the advisability of performing partial laryngectomy in the presence of fixation of the vocal cord, in a recent study Kirchner & Sam (1971) concluded that such a procedure could be performed in selected cases. From our study we think such a procedure ill advised, mainly because of the inaccuracy of the clinical assessment in these cases. This is particularly true following irradiation.

This study was supported by the Charlie Conacher Research Fund and the Ontario Cancer Treatment and Research Foundation (grant no. 707).

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MATERIAL AND METHOD

In the Toronto area virtually all patients with laryngeal carcinoma are referred to the Prin-

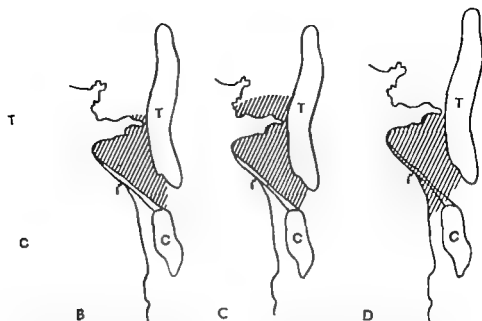


Fig 1 Histopathological classification. Illustrations depicting: (A) glottic tumour confined to the glottic region; (B) glottic tumour with minimal supraglottic extension; (C) glottic tumour with supraglottic extension; (D) glottic tumour with subglottic extension. C = cricoid cartilage. CE = cornu elasticum. T = thyroid gland.

also surrounding the laryngeal ventricle ("transglottic") and (D) glottic tumour with subglottic extension. C = cricoid cartilage. CE = cornu elasticum. T = thyroid gland.

Princess Margaret Hospital (a radiotherapy centre) for assessment before therapy. This assessment is carried out by a radiotherapist and a laryngologist working as a team. The methods of assessment include indirect and direct laryngoscopy (more recently including micro-laryngoscopy) supported by radiologic examinations (plane films, laryngeal tomography and a contrast laryngography). Most of these patients are treated by primary radiotherapy reserving operation for irradiation failure. However Bryce & Rider (1971) of Toronto recently studied a series of patients treated by

planned pre-operative radiotherapy followed by laryngectomy 6 weeks later.

This oncologic centre, the Princess Margaret Hospital, maintains careful and complete follow up records, making it ideally suited for retrospective studies. In order to assess the prognostic significance of vocal cord fixation we reviewed 134 patients from the Princess Margaret Hospital with $T_2N_0M_0$ glottic carcinomas (classified using the UICC criteria 1968). For this portion of the study those patients having limited mobility without complete fixation of the vocal cord are grouped together with those having complete fixation of the cord. The period of study covered 35 years from 1930 to 1965. Most of these patients were treated by primary radiotherapy and we have a minimum five-year follow up on all patients. Of these 134 patients, 61 had no vocal cord mobility and 73 had mobility of the vocal cord. In the group with normal mobility the crude five-year survival was 70%. In the group with impaired mobility the crude five-year survival

Table 1. T_2 glottic tumours—5 year absolute survival. All modes of therapy

	No of patients	No of survivors	survival
One response, normal mobility	61	43	70
One response, impaired mobility	40	25	63
One response, impaired mobility	33	15	45
			55



Fig 2 Spread outside larynx—thyro-arytenoid muscle invasion. Coronal section through a larynx at the level of the vocal process. The tumour (arrows) occupying the left glottic region, has extended subglottically and spread outside the laryngeal framework through the cricothyroid space with invasion of the upper margin of the cricoid cartilage (C). (Post irradiation.) FC=false cord. T=thyroid cartilage. C=vocal cord.

II Regional involvement Invaded structures

Distribution of tumour (histopathological)

Structure invaded	Confined to glottic region (1)	Glottic with supraglottic extension (1)	Glottic with subglottic extension (6)	Glottic with supra- and subglottic extension (20)	Total (28)
<i>Muscles</i>					
Thyro-arytenoid	1	1	6	20	28
Lateral crico-arytenoid			3	16	19
Interarytenoid		1	1	9	11
Posterior crico-arytenoid				7	7
Crico-arytenoid joint		1	1	15	17
Perineural space				10	10
Thyroid cartilage			1	15	16
Spread outside larynx				16	18



Fig. 3 Spread along muscle bundles. Horizontal section of a right partial laryngectomy specimen at the level of the vocal cord. The arrow marks the midline at the anterior commissure. The tumour has spread extensively along the muscle bundles of the thyro-arytenoid muscle beneath intact mucosa (postirradiation). C = cricoid cartilage. T = thyroid cartilage.

Figures confirm that fixation of the vocal cord is a poor prognostic sign in glottic carcinoma ($p=0.051$) (Table I).

In addition, we have had the opportunity of studying 110 laryngectomy specimens removed for carcinoma during the period 1966 to 1971. These specimens were examined by the method of whole organ serial sectioning (Tucker 1961 1963 Hyams, 1969). On the basis of the histopathology 73 of these 100 tumours were

considered to have arisen in the glottic area. The remaining 37 tumours were either supra glottic, subglottic, or multiregional. Of the 73 patients with glottic primaries, 28 were considered, on clinical assessment, to have fixation of the vocal cord at the time of laryngectomy. These 28 laryngectomy specimens form the basis for a detailed study of the causes of vocal cord fixation, and for a correlation between the clinical assessment and histopathological findings.

Of the 28 primary glottic tumours, histopathologically one was confined to the glottic region (Fig. 1 A) one had slight supraglottic extension (Fig. 1 B), six had slight subglottic extension (Fig. 1 D), and 20 had both subglottic and supraglottic extension. The descriptive term "transglottic" refers to neoplasms which cross the laryngeal ventricle and involve at least the glottic and supraglottic regions (McGavran et al. 1961) (Fig. 1 C). Seven of our 28 tumours could be described as having a "transglottic" distribution. In the histologic assessment of these tumours, we considered the conus elasticus to be the boundary between the glottic and subglottic regions, and the lateral angle of the laryngeal ventricle to be the boundary between the glottic and the supraglottic region (Fig. 1).

Of the 28 patients in this study 10 were treated by elective surgery 6 weeks after a full course of radiotherapy (5 500 rads over 6 weeks), 6 had elective surgery following 500 to 2 000 rads immediately pre-operatively 10 were treated by surgery following irradiation failure (including one partial laryngectomy) and the remaining 2 patients were treated by primary surgery.

In the irradiation failure group the length of time between the completion of irradiation and the surgical extenion varied from 4 to 33 months.

Causes of vocal cord fixation as histopathologically

In all 28 specimens the carcinoma invaded the thyro-arytenoid



Fig 4 Impaired vocal cord mobility (A) Coronal section of the larynx at the level of the middle third of the vocal cords. Tumour (outlined by arrows) occupies the left vocal cord and extends subglottically (S). Note the absence of invasion of the laryngeal framework. C = cricoid cartilage. T = thyroid cartilage.



(B) Photomicrograph of the same case showing nests of a moderately well differentiated keratinizing squamous cell carcinoma infiltrating the thyro-arytenoid muscle. There is extensive fibrous connective tissue surrounding the tumour foci (postirradiation).

r greater degree. This appeared to be the major factor in producing fixation of the vocal cord. The degree of involvement of the other intrinsic laryngeal muscles, the crico-arytenoid joint, and the thyroid cartilage depended upon the extent of the tumour. In some, there was also invasion of the perineural space. Many of the larger tumours had extended outside the laryngeal framework (Fig. 2). The details of the spread of these tumours are set out in Table II.

Invasion of the thyro-arytenoid muscle The degree of vocal cord fixation depends upon the extent of involvement of the thyro-arytenoid muscle. This muscle was invaded in all cases with fixation of the vocal cord. (In 4 additional cases in which vocal cord mobility

was limited without complete fixation, the tumour had only invaded the thyro-arytenoid muscle but had not extended to the deeper structures (Fig. 4).)

Tumour invasion of the thyro-arytenoid muscle has two distinct histopathological patterns. Many larynges including those who had salvage surgery for irradiation failure, had viable nests of infiltrating tumour scattered through the muscle (Figs. 7-8). In the remainder including the bulk of those treated by combined therapy there was extensive fibrous replacement of both the tumour and the muscle (Figs. 4-10). We also noted that once the thyro-arytenoid muscle had been invaded, the tumour tended to spread beneath intact mucosa along the length of the muscle bundles (Fig. 3).



B



D

Fig 3 Invasion of laryngeal muscles (B and D are from the same case, A and C from different cases).
 4) Coronal section through the tip of the vocal process showing tumour involving the thyro-arytenoid muscle and lateral crico-arytenoid muscle (LCAM) and lateral crico-arytenoid cartilage. E—ends prior to surgery) C—cricoid cartilage. E—

epiglottis. T—thyroid cartilage. (B) C through interarytenoid region shows interarytenoid muscle (IAM) and the cartilage (C) (postirradiation). T (C) Coronal section through the, showing invasion of the



Fig 6 Perineural invasion. Photomicrograph of circled area from Fig 5D showing perineural invasion.

Invasion of the other intrinsic laryngeal muscles. The next most frequently involved intrinsic laryngeal muscle was the lateral cricoarytenoid muscle (Fig. 5A) followed by the terarytenoid muscle (Fig. 5B). When this latter muscle was invaded the tumour often extended beneath intact mucosa to the opposite side. The posterior crico-arytenoid muscle (Fig. 5C) was infrequently involved and in each case the tumour was quite extensive.

Invasion of the crico-arytenoid joint. In 15 of the 28 specimens the crico-arytenoid joint space was directly invaded by viable tumour (Figs. 5C-8D). Two of the patients with

viable tumour in the joint also had an associated septic arthritis. In 2 additional cases the joint exhibited fibrous ankylosis suggesting that previously it had been involved by tumour. Both of these patients had been treated with combined therapy. Twenty-one patients had invasion of the arytenoid cartilage including all 17 of those with invasion of the crico-arytenoid joint.

Invasion of the perineural space. Invasion of the perineural space (Figs. 5D-6) was noted in ten specimens. Because the perineural space offers little resistance to tumour spread, tumour may travel considerable distances from

muscle (PCAS) and also invasion of the crico-arytenoid joint (postirradiation). A - arytenoid cartilage. C - cricoid cartilage. T - thyroid cartilage. (D) Coronal section through the arytenoid region showing extensive

invasion of the thyro-arytenoid muscle and cricoid cartilage (C). Circle indicates perineural invasion shown in Fig. 6 (postirradiation). C - cricoid cartilage. E - epiglottis. H - hyoid bone. T - thyroid cartilage

Table III Comparison between clinical assessment and pathological finding

Histopathological findings	Preoperative clinical assessment					
	Glottic T	Glottic with supraglottic extension T	Glottic with subglottic extension		Glottic with supra- and subglottic extension	
			T	T ₁	T	T
Confined to glottis					1	
Glottic with supraglottic extension					1	
Glottic with subglottic extension						
Confined to larynx	1		2		1	
Extending beyond larynx	1	1				
Glottic with supra- and subglottic extension						
Confined to larynx		4				
Extending beyond larynx		7	2	1	5	1

spread of the tumour. The same poor correlation (25% correct) was encountered if the clinical assessment of the mucosal extension of the tumour was compared with the pathology findings. Neither did our strict criteria for subglottic extension in the histopathological examination influence the poor correlation figures. This part of the study illustrates the difficulties involved in correctly assessing the real extension of laryngeal carcinoma.

DISCUSSION

Fixation of the vocal cord in glottic carcinoma has proved to be a poor prognostic sign (Heise & Baylis, 1966; Mårtensson et al., 1967; Vermond, 1970). This fact has recently been recognized by the UICC (Union Internationale Contre le Cancer, 1971) in its proposed alterations in the TNM classification of laryngeal carcinoma. In the present UICC classification developed in 1962 (UICC, 1968) fixation of the vocal cord in glottic T₁ tumours does not change the classification. In the proposed alterations, fixation of the vocal cord will automatically classify such a tumour as T₂. More over a glottic carcinoma localized to one vocal

cord but with fixation will also be included in this T₂ group (T₂ using current classification). Impairment of mobility will, however still be acceptable in the T₂ group. Similar alterations are anticipated by the American Joint Committee for cancer staging and end results reporting (AJC, 1971). Our study reinforces these proposals.

Fixation of the vocal cord which develops following the commencement of radiotherapy or after completion of radiotherapy is a particularly ominous sign. This development occurred in 5 of our patients, and in all, the fixation proved to be due to extensive viable tumour growth, sometimes beneath an intact mucosa (Fig. 8).

In glottic carcinoma the most important single factor in producing vocal cord fixation

Table IV Comparison between clinical assessment and pathological finding

Region involved	vertical extension	
Correctly assessed	8	29
Incorrectly assessed	20	
Spread outside larynx		15
deep extension		
Assessed	2	
Not assessed	15	

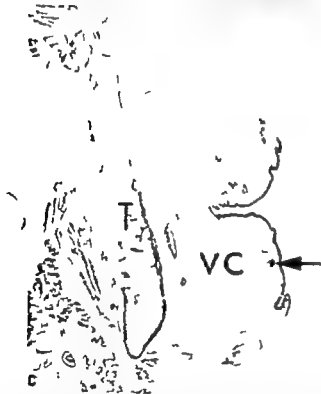


Fig 9 Partial laryngectomy—Impaired mobility (A, B, C and D are from the same case) (A) Gross photograph showing a partial laryngectomy specimen with an ulcerated tumour on the posterior portion of the vocal cord. C and D mark the coronal sections 9C and 9D (B) Photomicrograph showing a moderately well differentiated squamous cell carcinoma (post irradiation). (C) Coronal section (level C in 9A)

showing nests of tumour (arrow) beneath intact mucosa. FC=false cord. T=thyroid cartilage. VC=vocal cord. (D) Coronal section (through the visible tumour at level D in 9A) showing destruction of the vocal process by tumour and subglottic extension to the level of the upper border of the cricoid cartilage (C). T=thyroid cartilage

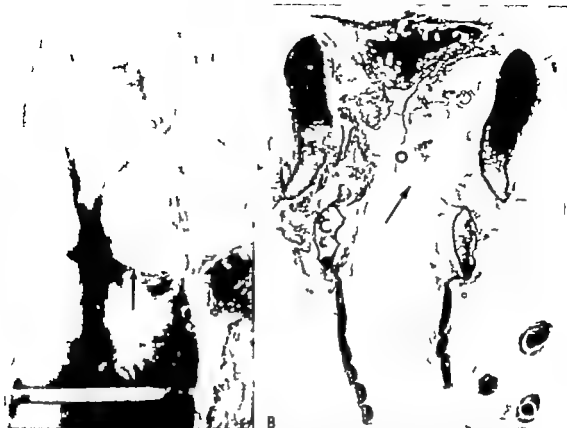


Fig. 18. Fixed vocal cord—partial laryngectomy post-mortem (A and B from the same case.) (A) Gross photograph of the larynx showing oedema of the right hemilarynx but intact mucosa. Arrow marks the level of coronal section 10 B (B) Coronal section showing

partial fibrosis of the thyro-arytenoid muscle. The corus elasticus is fragmented (arrow) suggesting earlier subglottic extension (postirradiation). Circled area (upper) showing nests of tumour (viable?). C = cricoid cartilage. T = thyroid cartilage.

direct invasion of the thyro-arytenoid muscle by tumour. This was present in all 28 specimens in this study (In an additional four cases the limitation of vocal cord mobility without complete fixation, the thyro-arytenoid muscle was the only structure invaded by tumour (Figs. 4 & 9).

Kirchner (1969) also found that "vocal cord fixation indicates deep invasion of the thyro-arytenoid muscle by cancer which often extends, and sometimes invades the thyroid gland.

When the invading tumour reaches the vocal folds of the larynx it tends to spread along the longitudinal axis of those muscle fibres (Fig. 3) presumably the route of least resistance. This feature becomes particularly impor-

tant in tumours involving the posterior commissure because they may extend beneath intact mucosa, across the midline, either in the submucosal layer or within the interarytenoid muscle (Fig. 5 B).

Invasion of the interarytenoid muscle has been noted as a cause of fixation by Kirchner (1969). In addition, in the review of 100 serially sectioned laryngeal carcinomas, Norris et al. (1970) found 10 carcinomas that invaded the interarytenoid muscle and 15 that invaded the space under this muscle.

Similarly invasion of the posterior crico-arytenoid muscle (Fig. 5 C), although it is infrequent (occurring in 7 of the 100 tumours), is particularly important for the surgeon because, in these cases, he

residual tumour in the pharyngeal mucosa of the postcricoid region. This feature has also been noted in other series (Kirchner 1969; Norris et al., 1970). Similarly Harrison in 1971 pointed out that if undetected, carcinoma will be left in the margins of the pharyngeal repair and the recurrence will be placed above the tumour.

In a high proportion of our patients (15 out of 28) the tumour directly invaded the cricoarytenoid joint space (Figs. 5C, 8D). In two more, the presence of fibrous ankylosis of the joint after irradiation therapy suggests previous tumour invasion. In their series of 100 serially sectioned specimens, Norris et al. (1970) reported 15 laryngeal carcinomas with joint invasion.

In the glottic carcinomas with intra- and subglottic extension, a high proportion invaded the thyroid cartilage and 16% spread outside the laryngeal framework, making wide field laryngectomy necessary (Figs. 7, 8).

We have been surprised by the extremely poor correlation between the clinical pre-operative assessment and the histopathological findings in our specimens. The high proportion of false-negative cases in this series, in part due to the very poor correlation between the clinical tendency of these lesions and the findings beneath intact mucosa, is in agreement with Gavran et al. (1959) conclusions that "it is common for a carcinoma to be clinically negative when examined histologically and radiologically appreciated". Similar discrepancies in the clinical assessment have been noted by Kirchner (1963) and may also be amplified by the pre-operative irradiation (Norris & Peale 1966; Goldman et al. 1966).

The faulty assessment of the tumours in the vertical plane (extent of involvement of supra-glottic and subglottic regions) despite the support of the radiological examination, further underlines the necessity of improvement in the diagnostic methods. These discrepancies have also been stressed by Olofsson et al. (1972).

Many surgeons believe that fixation of the vocal cord in glottic carcinoma is a contraindication for partial laryngectomy (Billler et al. 1970, 1971; Clerf et al., 1943; Kirchner 1969; Leonard & Litton 1971; Norris & Peale, 1966; Shaw 1966). Some surgeons, however, will consider partial laryngectomy on patients with limited vocal cord mobility (Billler et al. 1971; Sisson et al. 1970). Kirchner (1969) stated that "hemilaryngectomy and less extensive operations carry a high risk of inadequate removal of cancer in the case with a fixed cord".

More recently Kirchner & Som (1971) indicated that, in selected cases, cordal carcinoma with fixation might be amenable to hemilaryngectomy. They consider a subglottic extension of 8-9 mm or involvement of the inferior ventricular surface of the false cord renders the case unsuitable for partial laryngectomy. They emphasize that posteriorly the safety margin for subglottic extension is less. "The ultimate decision as to the feasibility of hemilaryngectomy in any and every instance must be made after exposure of the lesion by laryngofissure."

Partial laryngectomy was performed in one of our cases (Fig. 3). In two additional cases conservation surgery could have been performed instead of total laryngectomy if the surgeon had known the extent of the tumour at the time of surgery (Fig. 10). (In addition, two of our cases with limited mobility of the vocal cord had partial laryngectomy (Fig. 9).)

We believe that partial laryngectomy is contraindicated in most patients with glottic carcinoma and a fixed vocal cord. We base this conclusion mainly on the poor correlation between the preoperative clinical assessment and the pathological findings in these tumours, especially in cases with previous irradiation. We do not know how many of those cases reported by Kirchner & Som (1971) in their series of serially sectioned specimens had undergone irradiation before surgery.

It is often impossible to assess correctly the submucosal or extralaryngeal extension of

laryngeal carcinoma. We hope that the use of microlaryngoscopy in combination with improved radiological examination will give us more accurate clinical assessment in the future.

ACKNOWLEDGMENTS

I thank Usha Bhargava, who helped to collect the clinical information, Kenneth Ekem, Janice Elty and Alicia Tremmel in the Conacher Laryngeal Laboratory for preparing the slides, Harold Layne in the Department of Photography for his assistance with the photographic work, and Miss Gail Hyslop for typing this manuscript.

ZUSAMMENFASSUNG

Stimmbandcarcinome mit eingeschränkter oder aufgehobener Stimmbandbeweglichkeit haben eine schlechte Prognose.

Wir studierten 134 Fälle von T₁N₀M₀ Stimmbandcarcinomen. Von 73 Fällen mit eingeschränkter oder aufgehobener Stimmbandbeweglichkeit wurden in 40 Fällen 5-Jahresüberlebungen erzielt, in 55%. Von 61 Fällen mit normaler Beweglichkeit erhielten wir in 43 Fällen die 5-Jahresüberlebungsrate von 70%.

Außerdem wurden Serienschritte von 28 Kehlköpfen mit Carcinomen untersucht, die durch Laryngotomie entfernt worden waren. Der M₁ thyro-aryepiglottische Raum war in allen Fällen von Carcinomgewebe involviert. Die größeren Carcinome haben auch andere Räume wie andere Muskeln, die Art. crico-aryepiglottische und das knorpelige Gerüst angegriffen. Wir fanden schlechte Übereinstimmung zwischen klinischer Klassifizierung und histo-pathologischen Befunden. Mehr als 50% der Tumoren war klinisch präoperativ „unterklassifiziert“. Die Hauptursache dafür war Ausbreitung von Carcinomgewebe außen am knorpeligen Gerüst. Wegen der schlechten Möglichkeit einer richtigen präoperativen klinischen Klassifizierung hatten wir partielle Larynxektomie bei Stimmbandcarcinomen mit unbeweglichem Stimmband durchgeführt. Wir hoffen jedoch, daß verbesserte klinische und röntgenologische Untersuchungen in Zukunft bessere Möglichkeiten geben, Fälle für partielle Larynxchirurgie auszuwählen.

REFERENCES

the American Joint Committee (AJC) for cancer staging and end results reporting, 1971 *Clinical staging system for carcinoma of the larynx* (Corrected draft January 1971).
Biller H. F. Barash III, P. R. Ogura, J. H. & Perez,

C. A. 1970 Hemilaryngectomy following radiation failure for carcinoma of the vocal cords. *Laryngoscope* 80 249.
Biller H. F. Ogura, J. H. & Pratt, L. L. 1971 Hemilaryngectomy for T glottic cancers. *Arch Otolaryng* (Chic.) 91 38.
Bryce, D. P. & Rider W. D. 1971 Pre-operative irradiation in the treatment of advanced laryngeal carcinoma. *Laryngoscope* 81 1481.
Clarif L. H., Putney F. J. & O'Keeffe, J. J. 1948. Carcinoma of the larynx. *Laryngoscope* 53 632.
Goldman, J. L. Cheren, R. V. Zak, F. G. & Gimsberg M. J. 1966. Histopathology of larynges and radical neck specimens. In a combined radiation and surgery program for advanced carcinoma of the larynx and laryngopharynx. *Ann Otol* 75 313.
Harrison, D. F. N. 1971 The pathology and management of subglottic cancer. *Ann Otol* 80 6.
Hense, H. & Bayliss, P. 1966 Clinical staging in cancer of the larynx: a report on 788 cases using the American Joint Committee's method of stage classification. *Nat Cancer Inst* (US) 36 45.
Hyams, V. J. 1969 Personal communication.
Kirschner J. A. 1969 One hundred laryngeal cancers studied by serial section. *Ann Otol* 78 689.
Kirschner J. A. & Som, M. L. 1971 Clinical significance of fixed vocal cord. *Laryngoscope* 81 1029.
Leonard, J. R. & Litton, W. B. 1971 Selection of the patient for conservation surgery of the larynx. *Laryngoscope* 81 232.
Mårtensson, B., Fluor E. & Jacobsson, F. 1967 Aspects on treatment of cancer of the larynx. *Ann Otol* 76 315.
McGavran, M. H. Bauer W. C. & Ogura, J. H. 1961 The incidence of cervical lymph node metastases from epidermoid carcinoma of the larynx and their relationship to certain characteristics of the primary tumor. *Cancer* 14 55.
McGavran, M. H., Spjut, H. J. & Ogura, J. 1959 Laryngotomy in the treatment of laryngeal carcinoma. A critical analysis of success and failure. *Laryngoscope* 69 44.
Norma, C. M. & Peake A. R. 1966 Partial laryngotomy for irradiation failure. *Arch Otolaryng* (Chic.) 84 558.
Norma, C. M., Tucker G. F. Kuo B. F. & Pitzer W. F. 1970. A correlation of clinical staging, pathological findings and five year end results in surgically treated cancer of the larynx. *Ann Otol* 79 1033.
Olsson, J., Remouf J. H. & van Nostrand, A. W. F. 1972. Laryngeal carcinoma—correlation of radiography and histopathology (A study based on whole organ, serially sectioned laryngeal carcinoma specimens.) *Amer J Roentgenol* In press.
Shaw H. J. 1965 Glottic cancer of the larynx. *J Laryng* 79 1.
— 1966 Partial laryngectomy. *J Laryng* 80 839.
Shoen, G. A., Goldstein, J. C. & Becker G. D. Surgery of limited lesions of the larynx (in present). *Otol Clin North Amer* 3 529.
Tucker G. F. 1961 A histological

- study of the spread of carcinoma within the larynx. *Ann Otol* 70: 910.
- 1963 Some clinical inferences from the study of serial laryngeal sections. *Laryngoscope* 73: 728.
- Union Internationale Contre le Cancer (UICC) 1968. *T^NM classification of malignant tumours* (larynx classified 1962, pp. 22-24). Geneva.
- Union Internationale Contre le Cancer (UICC) 1971. *Classification of laryngeal carcinomas*. (Will be published 1972.) Personal communication. Delafressaye, J F Executive Director UICC, Geneva.
- Vermond, H 1970. Role of radiotherapy in cancer of the larynx as related to the T^NM system of staging. A review. *Cancer* 25: 485.

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REHABILITATION AFTER LARYNGECTOMY

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(Received August 21 1972)

abstract. Fifty patients have been assessed after laryngectomy 60% attained adequate vocal rehabilitation. Motivation was the principal factor determining success. Age and operative factors were found to have no influence on voice. Females were found to be less successful in this respect. 55% returned to their pre-operative employment, many to jobs requiring good voice or strenuous physical effort. Euphoria, subjective and objective was noted within 6 months after operation. The presence of the tracheostome was found to be troublesome to the majority of patients. Most patients suffered from post-operative depression for 6 to 12 months, mental stability was unaffected. Radical neck dissection was found to increase morbidity to a considerable extent.

Christian Theodor Billroth performed the first laryngectomy for cancer of the larynx nearly one hundred years ago in 1873 recurrence and death occurred within one month. Further attempts were performed over the ensuing years but cures were uncommon and mortality was high due to haemorrhage, sepsis, mediastinitis and bronchopneumonia (StClair Thomson 1939). Assessment of the number of laryngectomies performed in modern times is difficult, although Snidecor et al. (1962) estimated that there were 15 000-20 000 laryngectomies alive in the United States.

A substantial proportion of patients who undergo laryngectomy will have been cured of their disease and will die of other causes. The degree of rehabilitation achieved by these patients and their role in modern society is of importance. The problems of vocal rehabilitation have received much attention other

aspects of post-laryngectomy life have received less attention and assessments of overall rehabilitation are scant. The purpose of this paper is to make such an assessment.

METHOD AND MATERIAL

Fifty patients who had undergone total laryngectomy under the care of one of two surgeons at the Royal National Throat, Nose and Ear Hospital, and who were still living in the area were questioned, examined and tested by the author personally. No patients defaulted. Patients who had undergone pharyngo-laryngectomy were not included.

The series comprised 40 males and 10 females. Age distribution at the time of examination was as follows:

20-30 years	1 (female)
30-40 years	0
40-50 years	3 (1 female)
50-60 years	21 (3 females)
60-70 years	13 (2 females)
70-80 years	12 (3 females)

The youngest patient was a woman of 28 she had undergone surgery at the age of 2. The oldest was a woman of 78 she had undergone surgery at the age of 16. The oldest patient to have a laryngectomy was 14 at the time of operation.

The intervals between operation and examination were as follows:

3-6 months	4 patients
6-12 months	7 patients
1-5 years	19 patients
5-10 years	16 patients
10-18 years	4 patients

Four patients had signs of recurrent disease at the time of examination.

Ethnic origins

Anglo-Saxon,	47
Jewish,	1
Polish,	1
West Indian (negro),	1

Pre-operative visit by a laryngectomee

Twenty-six patients had been visited by a laryngectomee prior to operation. Of these, 18 considered this visit to have been of considerable value to them. The remaining 8 were indifferent, with the exception of one who found himself actively discouraged by the visit. Some who did not have a pre-operative visit felt that this would have helped them. Few patients were seen afterwards by a laryngectomee other than by the occasional chance meeting or by meetings at a laryngectomee club.

Radiotherapy

Twenty-eight patients had undergone a full course of pre-operative radiotherapy.

Operative details

All patients had Gluck Sorensen type incisions with lateral limbs where radical neck dissection was performed. Twenty-one cases had undergone radical neck dissection two of these being at a later date. No patients had undergone bilateral neck dissection. Operative notes, in the majority of cases, did not record details of reconstruction of the pharynx or cricopharyngeus muscle.

Pathology

Forty-nine cases had suffered from carcinoma of the larynx. The histology was squamous cell carcinoma with occasional cases being re-

ported as anaplastic carcinoma. One patient, a man of 45 had undergone operation for long-standing multiple papillomatosis.

Speech

An assessment was made of the patients' ability to communicate by oesophageal speech. Patients were divided into four categories falling into two groups:

Group A Adequate vocal rehabilitation. (i) Very good, superior speech resulting in fluent conversation with minimal intake gaps. (ii) Good, speech enabling conversation but with intake gaps.

Group B Inadequate vocal rehabilitation. (iii) Fair, a degree of oesophageal voice allowing some vocal communication, but not allowing conversation. Ability to make known needs in the home shops, etc. (iv) Poor, little or no oesophageal voice. Communication by whispered voice, signs and written word.

Results

Group A (i) Very good, 7 (ii) Good, 23 total 30 (60%).

Group B (iii) Fair 12, (iv) Poor 8 total 20 (40%).

Relationship of results to particular factors

1. *Age* Average age of patients in Group A is 61 years. Average age of patients in Group B is also 61 years. Seven (23%) of the patients in Group A were over 70 years of age (at the time of examination) and 5 (25%) in Group B were over 70 years of age.

2. *Sex* Three out of the 10 females fall into Group A.

3. *Nature of surgery* Twelve out of 10 in Group A had undergone radical neck dissection (45%) compared to 8 out of 20 patients in Group B (45%).

4. *Speech therapy* No relationship was evident between the number of attendances at speech therapy and the degree of oesophageal voice obtained. Many of the better speakers had had only few lessons.

5 Pharyngeal bar Thirty patients were noted to have a pharyngeal "bar" stretching from side to side across the pharynx at the base of the tongue (in a similar place to the normal epiglottis). In half these patients this bar was very marked. Eighteen out of the patients in Group A (60%) exhibited a pharyngeal bar. Twelve out of those in Group B had a bar (also 60%). One patient had a tight pharyngeal bar and complained at the time of examination of dysphagia. The bar was shortly afterwards divided under general anaesthesia. Recordings of his oesophageal voice before and after revealed no change in his voice after this procedure. His voice was considered good.

6 Radiotherapy No correlation was found between pre-operative radiotherapy and subsequent voice production.

Failures

An attempt was made to elucidate those factors which might have contributed to failure to attain adequate oesophageal voice.

1 Mrs N B., a 73-year-old woman (operated on at the age of 68), developed benign oesophageal stenosis requiring regular dilatation. She never achieved oesophageal voice.

2 Mr J R., a 75-year-old man (operated on at the age of 70) had previously had a cardio-vascular accident resulting in hemiplegia. His physical and mental condition was poor.

3 Mrs H. L., a 60-year-old woman (operated on at the age of 57) had a hiatus hernia.

4 Mrs G L., a 48-year-old woman (operated on at the age of 47), initially achieved reasonable oesophageal voice but did not exhibit oesophageal voice at the time of examination. It is probable that having attained voice she then rejected its qualities as being unfeminine.

5 Mrs R. W., a 28-year-old woman (operated on at the age of 25), never attained voice. It is probable that she also rejected the voice.

Failure in the remaining cases was more difficult to analyse. The overwhelming impression was gained that the principal factor was lack of motivation. Only one case who

failed to obtain voice expressed marked disappointment, this was case no 1 above in whom a definite organic cause for failure was found. The remaining patients appeared to accept their inadequate voice with comparative equanimity.

Discussion

The results of assessment of speech rehabilitation by various authors are variable. This is not surprising in view of the subjective nature of the problem.

Hayes Martin (1963) estimates that less than 50% rehabilitate adequately. Hunt (1964) reviewing 85 consecutive cases found 85.8% obtained "good" or "superior" speech. Murphy et al. (1964) reviewing 24 cases found 14 with adequate voice (58%). Robe et al. (1956) found 18 out of 32 to have adequate speech (56%). Putney (1958) reviewing 440 patients found a 62% success rate. The author's series with a success rate of 60% confirms the average findings of previous workers. There is no evidence in this series that age plays a great part in the degree of vocal rehabilitation. The figures suggest that females may be less successful in attaining good voice than males, although the figures are small. Putney (1958) however found two-thirds of the women in a series of 440 had inadequate voice and draws attention to the psychological factor involved. The low coarse tones of oesophageal speech tend to be distasteful and embarrassing to many women, there is no evidence that any other anatomical or physiological factor is involved.

The nature of the surgery does not appear to influence speech. Results were as good in patients who had undergone radical neck dissection as in those who had not. Putney (1958) also found no operative factors involved. He included consideration of skin incisions and the method of reconstitution of the pharynx. He found radical neck dissection to have no influence on voice.

There does not appear to be any relationship to the number of attendances therapy classes. The tendency is

those with poor or slow speech to attend classes longer than those who rapidly develop good voice.

Little mention is made in the literature of the pharyngeal bar exhibited in 60% of the patients in this series. The results, however, indicate that this bar is in no way related to the site of the pseudo-glottis.

The impression was gained that lack of motivation was the most important factor in the failure to obtain good speech. The introverted patient who is retired from work and lives alone sees little point in persisting if speech is not rapidly acquired. The patient with the over protective spouse may see little point in persevering (or occasionally may find it useful not to persevere). Some patients do not appear to exhibit the fortitude or industry necessary to acquire good speech. In only a handful of patients is there a possible pathological cause for failure.

The vast majority of patients will benefit from a visit by a "good" laryngectomee prior to operation: there are a few however who may be discouraged by such a visit, especially women. Post-operative visits, or early attendance at a laryngectomee club may be beneficial in many patients.

No patients in this series were using an α routinely: few had tried one. is the author's experience that the electro-larynx can improve communication for inadequate speakers even to the point of intelligible telephone conversation. The results of good oesophageal voice are, however, generally superior both from a practical and from a psychological point of view.

Symptoms attributable to aerophagia (flatulence, distention and borborygmi) occurred in some patients to a minor degree but in no case were they very troublesome.

One further aspect of speech deserves attention. Deterioration in speech is occasionally the first sign of recurrence of disease: usually a lymph node or mass of lymph nodes in the neck. This deterioration has been observed in patients before the patient was aware of the

recurrence and before the surgeon was able to detect it clinically. The mechanism of this early deterioration is not certain.

Occupation

Results

Forty patients were in regular pre-operative employment. Twenty-two (55%) returned to the same occupation. The remainder modified or ceased work as follows.

Modified, as a result of operation 5
 Changed, as a result of operation, 5
 Ceased, as a result of operation, 3
 Changed, reasons not directly related to operation 5

Of the 13 who had changed or ceased work as a result of the operation 5 stated their original job was too heavy. 3 were unable to climb heights, 3 changed because their voice was inadequate, one found the atmosphere too dusty (a wood machinist) and one (a commissionaire) could not comfortably wear a high collar.

Amongst those who returned to their original occupation some were particularly noteworthy:

- 1 Mr S C., a 59-year-old man continued to work as a lorry-driver.
- 2 Mr E. S., a 59-year-old man continued as a long-distance lorry driver.
- 3 Mr L. N., a 58-year-old man continued work as an information officer for an aircraft firm. His voice was in the very good category. He would initiate discussions with new clients by informing them why his voice sounded unusual.
- 4 Mr J. P., a 54-year-old man continued as a dock labourer for 5 years after operation.
- 5 Mr J. S. a 64 year-old man continued as a gardener.
- 6 Mr H. T. a 62 year-old man continued as a manual worker on the railways.

Discussion

It is clear that there are a number of occupational groups for which a laryngectomee is

susited. These do not necessarily fall into categories in which voice use or heavy vocal effort is at a premium. A number of have continued to perform jobs that red these attributes. In this series the ly occupational situations that appeared pletely to bar a laryngectomee were dusty spheres and heights. The former will include a wide variety of manual jobs. The latter includes workers on scaffoldings, buidlings and steplejacks. It is obviously unwise to employ people on such jobs who are not in prime physical condition.

It is perhaps surprising that only 3 patients found it necessary to change their occupations as a result of their voice. The degree of vocal rehabilitation achieved in some patients and the occupations that they continued to pursue is evidence that there are few occupations that cannot be followed by the successful oesophageal speaker if he so wishes.

There were no professional voice users in this series. It is obvious that such people (e.g. actors, singers and clergymen) will be unable to follow their professions in the manner to which they were accustomed.

The commonest stated reason for failure to return to work in the same capacity as before was that the work was too heavy. In some cases this may well have been a genuine inability to continue physically strenuous work as a result of the nature of this particular operation. In other cases there is little doubt that the opportunity to cease work was taken.

In those cases where the operation had forced a change or cessation of work, no cases of severe financial embarrassment were found, although financial loss occurred in some cases. It would appear that the Welfare State successfully protected patients in this respect. This series is selective in that it includes only those patients cared for within the State Medical Service. There may be a tendency for patients in higher socio-economic groups, in whom change of occupation may result in relatively greater financial hardship to seek private care.

The results indicate that the vast majority

of patients can, if they wish, return to full time occupations, often in circumstances which require regular voice use or strenuous physical effort although modification in the nature of the work may be beneficial.

Physical Capacity

Method and results

The physical capacity in forty-one patients was tested by lifting weights on a bar. Results were extremely variable: this is to be expected in a widely differing group of patients. The figures have no statistical significance and a control series was abandoned at an early stage. It may however be worth recording that 33 (80%) patients were able to lift a bar weighing 55 lb from the ground to a shoulder-high position using both arms. Twenty-three (56%) were able to lift the bar to an arms-extended-above-the head position. Twelve (30%) were able to repeat the latter procedure with a 75 lb bar.

Ability to lift weights of 35 lb or 55 lb with one arm to a waist-high position was assessed in 15 patients who had undergone radical neck dissection. In 10 of these patients the figures were the same on both sides. In the majority of these patients there was obvious difficulty in attaining the same result on the side which had undergone a neck dissection. In the remaining 5 patients, 4 could lift 35 lb weights on the normal side but could not lift this weight on the operated side: one could lift 55 lb on the normal side but only 35 lb on the operated side.

Many patients felt that their physical powers were reduced after operation. "Breathlessness" was a common complaint. Other patients took pride in being able to do as much after operation as before.

Discussion

The results suggest that a laryngectomee's physical capacity after surgery will not him from pursuing everyday activities: are however no indication that

capacity is not reduced by the nature of the operation. The extent to which lack of thoracic fixation affects physical power in man is still uncertain.

There is evidence to suggest that there is little change in physical powers after laryngectomy (Coyne et al., 1968). Sustained physical effort involves cardio-pulmonary function. There is no evidence that there is any significant change in these functions after operation. The complaint of breathlessness may indicate a greater awareness of the passage of comparatively cold, dry air in the tracheo-bronchial tree rather than an alteration in pulmonary function.

The results in patients who had undergone neck dissection suggest that physical capacity is limited on the operated side.

Further work in this field is indicated.

Olfaction

Method and results

Thirty-two patients stated that their sense of smell was poorer after operation. Eleven stated they had lost their sense of smell completely. Four noticed no change. Two had no sense of smell before or after operation. One noticed improvement after operation. Many patients had total anosmia for about 3 to 6 months after operation with some degree of recovery at about this time.

Tests were made using four substances in solution: lemon, cloves, peppermint and almond. Odour identification is unreliable and therefore odour perception was the only criterion used. Patients were tested as follows: (1) Solutions of the above substances were held in front of the anterior nares for up to 30 sec. (2) Those patients who were unable to perceive two or more of the test solutions were subjected to two insufflations of similar solutions into the nasal cavities from a Rogers insufflator.

Thirty-one patients (62%) were able to perceive two or more of the odours by method 1. Of the remaining 19, 17 (a further 34%) were

able to perceive two or more of the substances by method 2. Many of the patients who were able to perceive odours without insufflation into the nasal cavities were observed to perform inspiratory "sniffs" in the same manner as a normal person, although obviously the intake of air was through the tracheostome. This was apparently helpful in perceiving the odour. When the solutions were placed in front of the tracheostome there was no successful odour perception.

Discussion

The results confirm that decreased olfactory acuity is noticed by the vast majority of patients after laryngectomy. The increase in olfactory acuity on insufflation into the nasal cavities suggests that the hyposmia is simply due to diminished air-flow to the olfactory area. The majority of patients however were subjectively unable to perceive odours until at least 3 to 6 months after operation. Whilst an apparent anosmia might well be expected for some weeks after operation (in a patient who has more pressing problems to attract his attention) it is not clear why this should persist for as long as 6 months after operation. It may be that there is some alteration in olfaction during that time. It suggests that there may be a compensatory increase in olfactory acuity after operation but that such compensation takes some months to occur.

In some patients inspiratory "sniffs" appear to facilitate olfaction. It may be postulated that such movements, producing a negative intra-thoracic and therefore a negative intra-oesophageal pressure may in the absence of a normal cricopharyngeal sphincter result in the movement of a column of air which would result in a degree of air-flow into the nasal cavities.

Hyposmia after laryngectomy was reported by Henkin et al. (1968). They considered that the hyposmia was not related to air-flow; they found that gentle nebulization into the nasal cavities did not improve olfactory acuity. These findings are not confirmed in this series.

However all but six of Henkin's patients were tested within 6 months of surgery whereas 39 in this series were tested later than 6 months after operation. Henkin also commented on the subjective improvement in olfactory acuity at about 6 months in many patients.

A possible correlation was noted between the good oesophageal speakers and olfactory acuity 70% of the speakers with Group A voices were able to perceive two or more of the odours placed in front of the nares whereas only 35% of those in Group B achieved this success. This possible correlation may represent an increased ability to achieve air flow through the nasal cavities in the good speakers. It may however merely reflect the mental attitude of the patient who has achieved an overall greater degree of rehabilitation.

The author can offer no explanation of apparent increased olfactory acuity in one patient other than a psychological one.

The situation requires clarification and further pre and post-operative quantitative testing is indicated.

Respiratory Tract

Results

Nineteen patients complained of crusting in the tracheostome and trachea. Only 2 patients found this very troublesome. The remaining 29 patients had little trouble with crusting, although most had a certain amount in the first few months after operation. This later diminished. Cough, either dry or productive, was troublesome to a varying degree in approximately 30%. No patient gave a definite history of coryza and few felt that their susceptibility to upper respiratory tract infections was any greater after operation. Many patients had some degree of watery rhinorrhoea. On examination all patients exhibited the typical lilac coloured nasal mucosa associated with these patients.

Discussion

Despite the comparative paucity of complaints about the tracheostome with its associated

problems of cough, sputum and crusting, an impression was gained that these symptoms constitute a considerable problem to many patients. The fact that few complained markedly probably represents an acceptance of its inevitability rather than a lack of awareness of its presence.

Psychological Problems

Nearly all patients admitted to some early anxiety and depression. In most cases this lasted for about 6 months. Nine patients described this depression as being severe and lasting up to 12 months. A few received medication from their general practitioners but no patients had been referred for psychiatric treatment.

Harrison (1970) has stressed how different ethnic groups may react in differing ways to the operation. Some groups may regard the procedure as mutilating and may refuse operation or possibly commit suicide when they have discovered its effect.

Marital problems

The majority of patients felt that operation had caused little change in their marriage. In a few cases there was a feeling that the operation had brought the partners closer. One patient's marriage did break up. This was probably attributable to other causes. One patient felt that his condition had produced some degree of estrangement from his wife. Twenty-three patients had had regular sexual relations before operation of these, 19 were able to continue these relations after convalescence. Four did not continue their sexual relations after operation and attributed this, in part, to their operation.

It may be concluded that operation had no adverse effect on marital stability. Similar conclusions were reached by Murphy et al. (1964).

Effect of Radical Neck Dissection

Twenty-one patients had undergone radical neck dissection. All these patients

of symptoms attributable to this part of the operation namely pain and stiffness in the supra-clavicular and shoulder regions, inability to lift objects on the operated side and inability fully to abduct the arm. Some complained quite bitterly of these symptoms. The section on physical capacity above provides some evidence on this subject. There can be little doubt that a neck dissection adds considerably to the morbidity a morbidity that has perhaps been underestimated in the past. These facts should be borne in mind in considering prophylactic or elective neck dissections.

Worst Effect of Operation

Many patients had considerable difficulty in deciding what the worst effect of the operation was. The majority thought that the voice change was the worst effect and associated with this in many was the inability to laugh, sing or whistle. Some stated that the tracheostome was the most troublesome factor. Others considered the effects of a neck dissection to be their biggest problem. Sometimes it appeared that the worst effects were those that the patient had been least prepared for.

Conclusions

There can be little doubt that total laryngectomy imposes a considerable physical handicap. It requires changes in way of life and alters outlook. In the same way that people may overcome other physical handicaps to a remarkable degree so may the laryngectomee. This requires courage and stamina, especially in the early stages. A large number of these successful laryngectomees are proud of their achievements and rightly so. Given explanation, encouragement and optimism the laryngectomee can return to a full useful and enjoyable life.

ACKNOWLEDGMENTS

The author is grateful to Professor D. F. N. Harrison for encouraging him in this project and for allowing

him to examine his patients, to Mr Anthony Rad, for allowing him to examine his patients, to Mrs Joyce L. Cook, Senior Speech Therapist for helpful advice and to Miss V. Johns, Head Social Worker for advice and information.

ZUSAMMENFASSUNG

50 Patienten nach einer Laryngektomie wurden beobachtet. 60% erlitten angemessene soziale Rehabilitation. Motivation war der Hauptgrund für den Erfolg. Alter und operative Fakten hatten keinen Einfluss, allerdings waren die Ergebnisse bei Frauen weniger erfolgreich. 55% der Patienten nahmen Beschäftigungen wieder auf die sie vor der Operation ausübten hatten, viele im Beruf, die eine gute Stimme oder anstrengende körperliche Betätigung verlangten. Geruchlosigkeit, subjektiver oder objektiver Art, wurde bemerkt, jedoch verbesserte sich dies oft 6 Monate nach der Operation. Die Gegenwart der Tracheostoma wurde von den meisten Patienten als belästigend empfunden. Ein Grossteil der Patienten litt während 6-1 Monaten unter postoperativen Depressionen, die teilweise der Eben blieb jedoch unbefriedigend. Rallies Halsdissektion (Zergliederung) wurde als besonders sterblichkeitsvermehrend empfunden.

REFERENCES

- Coyne, J. M., Strum, J. R., Payton, O. H. Klein, G. A. & Arensler, J. F. 1965. The laryngectomee and lifting. *Arch. Otolaryng. (Chic.)* 83 106.
Harrison D. F. N. 1970. Personal Communication.
Henkin, R. I., Hoyer, R. C., Ketchum, A. S. & Gould, W. J. 1962. Hypoxemia following laryngectomy. *Lancet* ii 479.
Hunt, R. H. 1964. Rehabilitation of the laryngectomee. *Laryngoscope* 74 382.
Martin, Hayes. 1963. Rehabilitation of the laryngectomee. *Cancer* 16 823.
Murphy, G. E., Biano, D. L. & Ogura, J. H. 1964. Determinants of rehabilitation following laryngectomy. *Laryngoscope* 74 1533.
Petry, F. J. 1938. Rehabilitation of the post-laryngectomized patient. *Ann. Otol.* 67 344.
Robe, E. V., Moore, P., Andrews, A. H. & Hilder, P. H. 1956. A study of the role of certain factors in the development of speech after laryngectomy. *Laryngoscope* 66, 173.
Snideron, J. C. et al. 1962. Speech rehabilitation of the laryngectomized. Charles C. Thomas, Springfield, Ill.
St. Clair Thomson. 1939. The history of cancer of the larynx. *J. Larynx* 54 61.

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STIMULATION OF SYMPATHETIC NERVE FIBRES TO THE NOSE IN CATS

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(Received September 18 1972)

Abstract The sympathetic innervation to the nose of the cat has been investigated by two methods. Venous flow from one nose cavity has been recorded as drops of blood and simultaneously the nasal patency from the same cavity has been estimated by a balloon technique. More than one sympathetic pathway giving decreased venous flow and increased nasal patency in the nose cavity has been demonstrated. Some of the sympathetic postganglionic nerve fibres travel in the vidian nerve together with parasympathetic preganglionic nerve fibres. When the vidian nerve was stimulated with low intensities and high frequencies the parasympathetic effects dominated. With high intensities and low frequencies, the sympathetic effects dominated. In one of 17 cats no sympathetic fibres were found in the vidian nerve. Other sympathetic pathways to the nose are discussed. Finally stimulation of the contralateral cervical sympathetic nerve affects the nose cavity

Vasomotor rhinitis is supposed to be caused by an imbalance between sympathetic and parasympathetic impulses to the vessels and glandular elements of the nasal mucosa with a relative overactivity in parasympathetic nerves during attacks (Golding Wood, 1961). The pathways of the autonomic nerves to the nose in man are not known precisely but are thought to be the same as in certain animals of which the cat has been most thoroughly investigated.

The parasympathetic preganglionic supply to the nose emerges from the brain stem with the facial nerve but after a short distance leaves it as the greater superficial petrosal nerve. This nerve then enters the pterygomaxillary

fossa and there the preganglionic fibres synapse with the postganglionic in the pterygopalatine ganglion. Through the pterygopalatine foramen the postganglionic nerve fibres travel to the vessels and glandular elements in the nasal mucosa.

According to textbooks in anatomy the sympathetic postganglionic nerve fibres from the superior cervical ganglion travel along the internal carotid artery leave it as the deep petrosal nerve then join the parasympathetic nerves in the pterygoid canal and accompany these to the nasal mucosa.

An exception to this distribution of the parasympathetic and sympathetic pathways was shown by Malcomson (1959) who found the ethmoturbinal area of the nose in the cat innervated via the ciliary ganglion.

The nerve in the pterygoid canal and in the pterygomaxillary fossa central to the ganglion is called the vidian nerve and as described above it consists of preganglionic parasympathetic and postganglionic sympathetic nerves.

The rationale to treat vasomotor rhinitis would be to cut the parasympathetic supply only but this requires intracranial surgery. Golding Wood (1962) and other authors have, however, made a vidian neurectomy that they have also cut the sympathetic nerves; their results are described as good. The reason for this? Is there a good

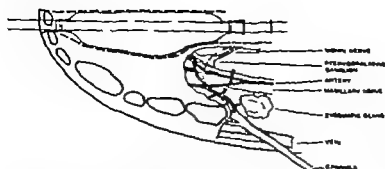


Fig 1 A schematic drawing of the transpalatal approach to the vidian nerve and the vein from the pterygopalatine foramen. The vein is marked with oblique lines and the artery is black. The nerves are trifurcated.

tion of the sympathetic nerves and poor regeneration of the parasympathetic ones? Are there many pathways for the sympathetic supply to the nose? Are there any sympathetic fibres at all in the vidian nerve?

Malcomson (1959) has made experiments on cats indicating that the sympathetic supply reaches the nose via the vidian nerve Jackson & Rooker however have recently (1971) called in question whether a vidian nerve section would remove a significant portion of nasal sympathetic fibres at least in dogs. These authors have used similar recording techniques and have measured the pressure necessary to force a flow of air through the nose.

In the present investigation a new method is described, with which sympathetic pathways to the nose of cats were studied. The method implies that most of the venous flow from one of the nose cavities is measured in combination with pressure changes in a water filled balloon in the same cavity.

METHOD

The results in this report were obtained from 24 cats. The animals were given chloralose anaesthesia (80 mg/kg intravenously) after induction with ether. Additional doses were given when required. Breathing was ensured by inserting a tracheal cannula. The dissection of the pterygomaxillary fossa started with an incision in the buccal mucosa near the innermost tooth in the upper jaw. Under the mucosa a branch of the facial vein could be identified and traced just behind the tooth. Then the

mucosa of the soft palate was deflected from the tooth to near the midline. The pterygoid muscle was removed carefully and under it the vidian nerve was seen and dissected free from adjacent tissue. The pterygopalatine ganglion was often concealed by the hard palate and in these cats some of the bone had to be drilled away. The vein was followed under microscope in retrograde direction to the pterygopalatine foramen. On the way to the foramen 7 to 10 branches of the vein were ligated, some of them much larger than the dissected vein. Fig 1 is a drawing of the dissected region. A small water filled balloon was inserted in the nose cavity on the dissected side. The balloon consisted of a very thin latex tube with a diameter of about 8 mm tied at both ends over a polyethylene tube with an outer diameter of 1.6 mm. The length of the tied balloon was about 20 mm. The polyethylene tube had small holes within the balloon. The function of the polyethylene tube was to make the insertion into the nose cavity easier and to conduct the pressure changes in the balloon to a transducer and a polygraph. The system was closed but the resting pressure in the balloon could be set at any desired level from a pressure bottle. Except in some experiments the level was set to 6 cm of water above the nose cavity. The cervical sympathetic nerve on the same side was prepared and sectioned and then heparin was given. The arterial pressure was recorded in the femoral artery with a mercury manometer. The vein from the nose cavity was cannulated with a polyethylene tube of widest possible bore as indicated in Fig 1 and the venous blood was conveyed to a drip

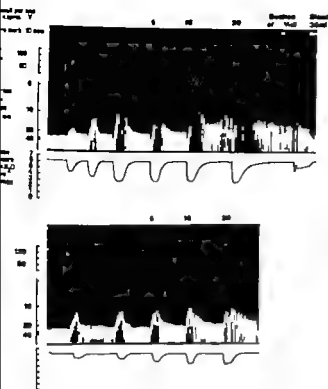


Fig. 2. Cat, 3.2 kg. Responses to stimulation at various frequencies of the cervical sympathetic nerve before and after the vidian nerve has been cut. Stimulation periods and injections are marked underneath time record. Lowest record represents balloon pressure manually transposed from the polygraph record. A decreased balloon pressure marks an increased nasal patency.

number with a photocell connected to an automatic writer recording on a smoked drum. The blood was returned to the cat by intravenous injections.

The peripheral end of the cervical sympathetic nerve was stimulated with a unipolar electrode. Square-wave pulses of an intensity of 4 V and a duration of 3 msec and of various frequencies were used. The peripheral end of the vidian nerve was placed on a bipolar electrode and the distance between the two points of contact was about 1 mm. The duration was 3 msec and the intensities and frequencies were varied as described later.

After the experiment had been done and the cat had been anaesthetized by air intravenously, the nasal cavity was fully opened via the hard palate. Methylene blue solution was then injected in retrograde direction through the catheter in order to mark out the region sending blood from the nose.

Hexamethonium bromide and dihydroergotamine methanesulphonate were administered intravenously.

RESULT

Stimulation of the cervical sympathetic nerve before and after section of the vidian nerve

The resting venous flow from one nose cavity varied between 10 and 30 drops per min in all cats, that is 0.5 to 1.5 ml per min. With an intensity of 4 V and with various frequencies the peripheral end of the cervical sympathetic nerve was stimulated in 11 cats. The nasal patency increased and the maximal responses were generally reached at 10 impulses per sec. Simultaneously the venous flow decreased, as can be seen in one example in Fig. 2. The responses in both records were larger before than after section of the vidian nerve, indicating that some of the sympathetic nerve fibres travel in the vidian nerve. But in all 11 cats there were marked responses when the vidian nerve had been cut, indicating that there are other ways to the nose other than the vidian nerve. As can be seen in Fig. 2, the responses before and after

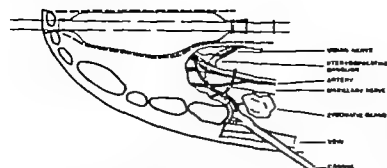


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did not increase the nasal patency as also evidenced in Fig. 3. The effect on the venous flow was investigated in only 3 of the 8 cats. Stimulating the contralateral cervical sympathetic nerve gave a small but an obviously decreased venous flow.

stimulation of the vidian nerve with various intensities and frequencies

The peripheral end of the sectioned vidian nerve was electrically stimulated in 17 of the cats. In 13 the intensities were varied from 2 to 12 V and in 5 of these the frequencies from 1 or 1 to 20 per sec. The effects of stimulating the vidian nerve with a constant frequency of 10 per sec and with various intensities can be seen as mean values (with S.E.M.) in Fig. 4.

The nasal mucosa never reacted at 2 V but as a rule at 3 V. From 3 to about 8 V the venous flow was faster than the resting flow with a maximum at about 4 V. From about 8 to 2 V the venous flow decreased on stimulation. The nasal patency decreased from 3 to about 4 V and over 4 to 5 V it increased. Thus the reversal for the venous flow occurred at higher intensities than the "reversal" for the nasal patency. In order to investigate why the

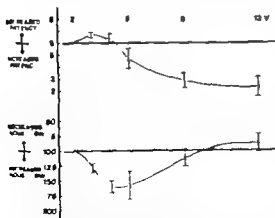


Fig. 4 Responses to stimulation of the vidian nerve at a frequency of 10 sec and at various intensities. Mean values with standard error of mean from 13 cats. Ordinates from above: pressure in the balloons in cm of water above the nose, changes of the venous flow as percentage of the resting flow. Abscissa: stimulation volts.

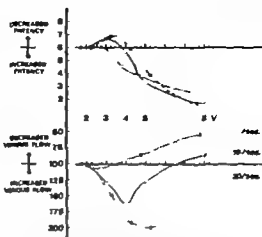


Fig. 5 Responses to stimulation of the vidian nerve at various intensities and frequencies. Mean values from 5 cats. Ordinates and abscissas as in Fig. 4.

two "reversals" were obtained at different intensities the outflow level of the falling venous drops and the resting pressure in the balloon were varied, but this made very little difference.

In one cat there was no sign at all of vasoconstrictor nerves in the vidian nerve.

In 5 cats both frequency and intensity were varied on stimulation of the vidian nerve. In all of them the same regular patterns could be seen for the venous flow. At low frequencies, for instance 0.5 and 1 per sec, the venous flow was lower than at 10 and 20 per sec at constant intensities. Stimulation at low frequencies gave, as a rule even slower flow than at rest except below 4 V where it gave an increased venous flow. This indicates that the threshold for stimulation of the postganglionic sympathetic nerve fibres in the vidian nerve at a duration of 3 msec and with the electrodes used lies at about 4 V. The mean values of the 5 cats are given in Fig. 4 and as can be seen, the nasal patency did not change as regularly as the venous flow.

Stimulation of the vidian nerve before and after a ganglionic blocking agent

After stimulating the vidian nerve electrically 4 cats were given 5 mg/kg hexamethylenetetramine.

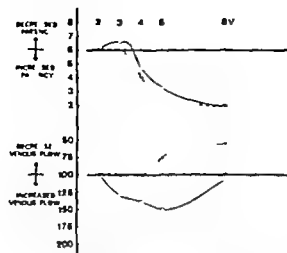


Fig 6 Responses to stimulation of the vidian nerve at 10 sec and various intensities before (—) and after 5 mg/kg hexamethonium intravenously. Mean values from 4 cats. Ordinates and abscissae as in Fig. 4

intravenously. The immediate effect of this ganglionic blocking agent was a fall of the blood pressure, a decrease of the venous flow and an increase of the nasal patency in all the cats. Five to 15 min later the nerve was again electrically stimulated at intensities from 2 to 8 V and frequencies from 0.1 to 20 per sec. The results were a decreased venous flow and an increased nasal patency. The differences before and after hexamethonium were largest at 10 to 20 per sec and least at low frequencies. Mean values of the 4 cats stimulated at a frequency of 10 per sec and at various intensities are given in Fig. 6.

Stimulation of the vidian nerve before and after an α -adrenergic blocking agent

Dihydroergotamine, a drug with α -adrenergic blocking effects, was given intravenously in doses of 0.1 to 0.5 mg/kg, sometimes 2 to 3 times to the same cat. As a rule the blood pressure fell markedly at the first dose and then rose very slowly for one-half to one hour to (for the experiment) acceptable values. When the blood pressure decreased the nasal patency always increased. The venous flow on the other hand, did not always decrease. In some cats it did not change very much. Owing

to the pronounced fall of the blood pressure, results were obtained from 4 cats only. The vidian nerve was stimulated with a frequency of 10 per sec and with intensities between 2 and 12 V. After dihydroergotamine the venous flow was higher for intensities over 4 V. The nasal patency decreased but not always for the highest intensities. The "reversal" for the nasal patency had not disappeared but changed place as a sign of incomplete sympathetic blockage. Mean values of the 4 cats are given in Fig. 7.

DISCUSSION

With methods reported in the earlier literature it has only been possible to measure the degree of nasal patency. A decreased nasal patency has been designated as vasodilatation and an increased as a vasoconstriction, but in this article it is preferred to avoid the terms of vasodilatation and -constriction. As we know that the system of vessels in the nose is very complicated and that there are large reservoirs for blood in the sinusoids, it is clear that changes in nasal patency and blood flow through the nose do not necessarily run parallel. The two methods described above offer the possibility to measure these changes simulta-

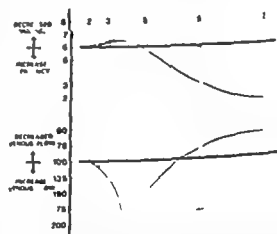


Fig 7 Responses to stimulation of the vidian nerve at 10 sec and various intensities before (—) and after dihydroergotamine in doses of 0.1 to 0.5 mg/kg intravenously. Mean values from 4 cats. Ordinates and abscissae as in Fig. 4

taneously. But a comparison can be made only qualitatively and not quantitatively because there is no evidence that the collecting vein drains just the same part of the nasal mucosa as that covered by the balloon.

As early as in 1913 Tschalussow demonstrated decreased nasal patency after electrical stimulation of the vidian nerve in the cat and since then the existence of vasodilator nerve fibres to the nose has never been denied. The existence of nerve fibres in the vidian nerve giving an increased nasal patency has, however, been more debated. Malcomson (1959) needed intensities of stimulation of more than 7 to 10 V to get an increased nasal patency in cats and Jackson & Rooker (1971) have suggested that such high intensities may give an irradiation to sympathetic fibres along vessels and sensory nerves in the vicinity. The latter authors, working on dogs, found no or very few sympathetic fibres in the vidian nerve.

In this investigation the responses to stimulation of the cervical sympathetic nerve were significantly smaller after than before section of the vidian nerve, and just when the latter nerve was cut a brief reduction of the venous flow and an increase of the nasal patency appeared. These results, together with the effects of direct electrical stimulation of the vidian nerve with and without a ganglionic and an α -adrenergic blocking agent, tend to prove the existence of sympathetic fibres in the vidian nerve in the cat. But as the responses to stimulation of the cervical sympathetic nerve after the vidian nerve was cut were very obvious in all the cats, other sympathetic pathways must exist. It is not possible, however, with the present methods to estimate the proportions of sympathetic fibres in the different pathways. Furthermore, the proportions of sympathetic nerve fibres in different pathways do not seem to be the same from cat to cat, best exemplified by one of the cats in which no sympathetic nerve fibres at all could be demonstrated in the vidian nerve.

Sympathetic pathways other than in the vidian nerve have been proposed and demon-

strated before. Malcomson (1959) stimulated nerve fibres central to and peripheral to the ciliary ganglion and got an increased nasal patency. Only a small part of the nose, the ethmoturbinal area, is supposed to be innervated by these nerves. The blood to this area comes from the ethmoidal arteries and the venous blood probably leaves by adjacent vessels and not by the vein passing through the pterygopalatine foramen. Therefore, stimulation of nerve fibres passing or relaying in the ciliary ganglion probably does not affect the venous flow in the present experiments. Another and certainly not unimportant sympathetic pathway to the nose may be along the external carotid artery as proposed by Jackson & Rooker (1971).

Finally impulses in sympathetic nerve fibres from the contralateral side have an effect on the nasal patency. There is probably an effect on the venous flow too: the results from 3 cats point in that direction. Whether the sympathetic nerves cross the midline is not known.

If these results can be transferred to man there seems to be, after sectioning of the vidian nerve and without regeneration, enough sympathetic nerve fibres to the nose to give a good vasoconstriction or increased nasal patency and this might explain some of the good results in vasomotor rhinitis after vidian nerve section.

Sympathetic nerve endings are richly represented around the veins of the nose in rat, guinea-pig, rabbit and cat (Dahlström & Fuxe, 1965). An indication of the presence of sympathetic nerves to the veins is given in the present experiments by the very early brief increase of venous flow after electrical stimulation of the cervical sympathetic nerve. After this increase, the prolonged decrease followed. During the whole stimulation, even in the early stages, nasal patency was increased. The early brief increase in venous flow was more often seen on cervical sympathetic stimulation before than after the vidian nerve had been cut. But this can hardly be taken as evidence that sympathetic nerve f

veins prefer the vidian nerve to other pathways.

The effects on the nasal mucosa of stimulating the vidian nerve with various intensities and frequencies seem to be due to at least two important factors. One is that with growing intensities within certain limits, more and more nerve fibres of both kinds are activated. The other is that the two sets of autonomic nerves have different frequency and intensity-response curves. This has of course been obvious to other investigators but from the present experiments with a ganglionic and a sympathetic blocking agent it has now been demonstrated. The threshold for stimulating parasympathetic nerve fibres is about 3 V and more and more fibres are activated up to 4 to 5 V. The threshold for sympathetic fibres is about 4 V but even up to 8 V more and more fibres seem to be stimulated. At low frequencies of 0.1 to 1 per sec the sympathetic effects are dominant and at 10 to 20 the parasympathetic effects dominate.

ZUSAMMENFASSUNG

Die sympathische Innervation im Nasen von Katzen wurde mit zwei Methoden untersucht. In einem Nasenraum wurde die venöse Strömung durch die Blutropfen registriert und gleichzeitig das An- und Abschnellen der Nasenschleimhaut mit einem eingeklemmten Ballon gemessen. Dabei stellte sich heraus,

dass mehrere sympathische Bahnen vorhanden sind, die bei Reizung die venöse Strömung vermindern und die Nasenschleimhaut abschnellen lassen. Ein Teil der sympathisch-postganglionären Nervenfasern läuft im Nervus Vidianus mit parasympathisch-präganglionären Nervenfasern zusammen. Bei Reizung des Nervus Vidianus mit schwacher Intensität und hoher Frequenz dominiert der parasympathische Effekt, dagegen bei hoher Intensität und niedriger Frequenz der sympathische. Bei 17 Versuchen war eine Katze, bei der keine sympathische Faser im Nervus Vidianus gefunden wurde, die einzige Ausnahme. Weitere sympathische Bahnen der Nase werden diskutiert. Reizung der kontralateralen zervikalen sympathischen Nerven beeinflusst die Nasenschleimhaut.

REFERENCES

- Dahlström, A. & Fuxe, K. 1965 The adrenergic innervation of the nasal mucosa. *Acta Otolaryng* (Stockh.) 59 88.
Golding Wood, P. H. 1961 Observations of pre- and vidian neurectomy in chronic vasomotor rhinitis. *J Laryng* 75 23.
— 1962. Pathology and surgery of chronic vasomotor rhinitis. *J Laryng* 76 969.
Jackson, R. T. & Rooker D. W. 1971 Sympathetic and section of the vidian nerve in relation to autonomic control of the nasal vasculature. *Laryngoscope* 81 565.
Malcomson, K. G. 1959 The vasomotor action of the nasal mucous membrane. *J Laryng* 71 73.
Tuchatschnow M. A. 1913 Die Innervation der Gefäße der Nasenschleimhaut. *Pflüger Arch Ges Physiol* 151 523.

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ON THE ORIGIN OF LYMPHOID CELLS IN THE PALATINE TONSIL

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(Received September 18, 1972)

Abstract The origin of the lymphoid cell populations of the tonsil was studied by sublethal whole body irradiation of normal and thymectomized animals. Two clearly distinguishable cell populations appeared to be present in the tonsil: a thymus-dependent population, situated in the interfollicular areas, and another presumably bone marrow derived lymphoid cell population, localized in the follicular structures. From the experimental data obtained, it is concluded that no fundamental difference exists between the lymphoid tissue of the tonsil and that of the regional lymph nodes in the adult animal.

In the immunological response two different elements can be distinguished dependent on the nature of the antigen: the plasmacellular reaction (humoral immunity) in which specific antibodies are formed and the specific cellular reaction (cellular immunity) in which committed lymphocytes are involved.

The course of the plasmacellular reaction has been described by Fagraus (1948). She demonstrated that antigenic stimulation gives rise to large basophilic blast cells, which are transformed into mature plasmacells. During this process a sharp rise in circulating antibodies, produced by these cells was observed. The origin of the precursor cells of this reaction is in the bone marrow as has been demonstrated by Balner & Dersjant (1964), Miller & Mitchell (1968), Nossal et al. (1968) and Miller & Mitchell (1969).

The specific cellular reaction is also ac-

companied by the formation of large basophilic cells but transformation in plasma cells does not occur. The end cells of this reaction are small lymphocytes (Gowans et al., 1963; Oort & Turk, 1965). These cells are involved in the cell-mediated immune reactions such as homograft rejection, delayed hypersensitivity and sensitization by chemical substances like DNCB (Gowans et al., 1963; Keuning, 1965; Turk & Oort, 1969). It has been shown that mature thymocytes are the immunologically component cells in the specific cellular immune response (Miller 1962; Keuning & van den Broek, 1968; Veldman 1970).

In recent years experimental procedures have been developed facilitating the separation of these two elements of the immune response. Keuning & van den Broek (1968) subjected adult rabbits to repeated sublethal X-irradiation to eliminate temporarily nearly all lymphoid cells. To prevent the regeneration of the thymus-derived population of lymphoid cells, the animals were thymectomized before irradiation. In these animals the restoration of the antibody-forming capacity simultaneous with the reappearance of the follicular structures was observed 7-8 days after the last irradiation. However, no repopulation of the paracortical areas in the regional lymph node occurred, while the potency for homograft rejection was abolished throughout an observation period of more than 100 days. Comparable experiments in which thymus was applied, no significant loss of

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was observed in the paracortical areas of the regional lymph nodes, while the capacity for homograft rejection was not affected (Veldman 1970). The restoration of the antibody-forming potency was the same as in the thymectomized animals. In this way a clear morphological and functional separation between the two different elements in the immune response could be obtained.

On the role played by the palatine tonsil in the immunological system there is a large diversity of opinions. Several hypotheses have been put forward, as for instance that the tonsil is an antibody-producing organ (Meyer zum Gottesberge & Koburg, 1967; Surján & Surján 1970) or an equivalent of the central lymphoid organs as the thymus (Harrison, 1970) or the bursa of Fabricius (Fichtelius, 1968).

The present report deals with an experimental study on the tonsil in which the origin of the lymphoid cell populations was traced with the use of the above-mentioned techniques and thus to obtain more insight into the role played by the tonsil in the immunological defense mechanism.

METHOD

The experiments were performed on adult white rabbits of the New Zealand strain (about 1 kg body weight). For whole body irradiation a source of radioactive Cobalt⁶⁰ was used. During irradiation the animals were fixed in a wooden box. A sublethal dose of 450 rads was administered three times at biweekly intervals. The radiation dosage was measured in the midline of the animal and administered in two equal amounts, one half to the left and the other half to the right side of the body. Thymus shielding was performed with the aid of lead blocks.

For thymectomy endotracheal anesthesia was given, using a closed system through which a mixture of halothane nitrous oxide and oxygen was administered. Artificial respiration was started after opening the thorax

by severing the first two ribs from the sternum. The exposed large thymus lobe was removed from the mediastinum by blunt dissection as well as the small cranial extensions along the carotid arteries. Subsequently the ribs, the muscular layer and the skin were sutured. Irradiation of these animals was started about 4 weeks after the surgical procedure.

At various times after the last irradiation the animals were killed by intravenous injection of a lethal dose of Nembutal. In the thymectomized animals the thymus region was carefully inspected for the presence of thymus remnants. In none of the animals was any thymus remnant observed. Tonsils and popliteal lymph nodes were dissected, fixed in Zenker's solution and embedded in paraffin wax. Serial sections (7 μ m) were stained with methylgreen-pyronine.

RESULT

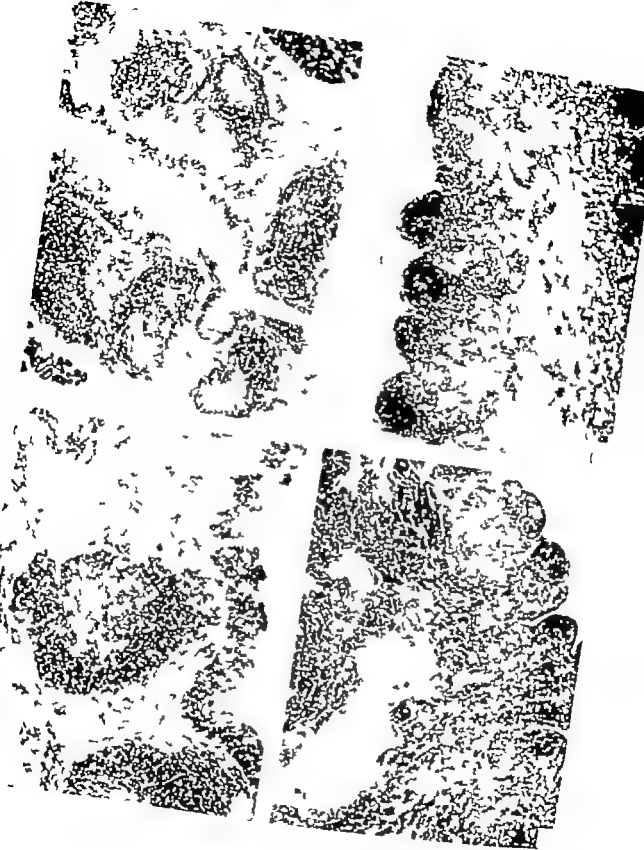
Micrographs of the structure of the normal palatine tonsil and popliteal lymph node of a young adult rabbit are shown in Figs. 1 and 2. The tonsil consists of a simple epithelial crypt. The epithelium is usually infiltrated to a varying degree by phagocytotic and lymphoid cells. Just below the epithelial layer a single row of follicles is arranged, with their lymphocyte corona directed towards the epithelium. This corona consists mainly of small dark-staining lymphocytes with scattered phagocytotic cells. The germinal centres, situated below the corona and revealing many mitotic figures, are populated by medium sized lymphocytes, blast type cells and phagocytotic cells. Between the follicles large accumulations of small lymphocytes and many epithelioid venules are observed. On its outer side the tonsil is surrounded by a large mucous gland. Normally the ducts of this gland open into the pharynx, but sometimes openings are found into the crypt (Fig. 3). The popliteal lymph node shows the normal characteristics of regional lymph nodes with a central marrow space, surrounded by follicles and the so-called paracortical areas

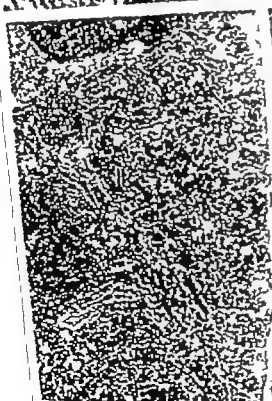


Figs. 1-2 Tonsil (Fig. 1, 60) and popliteal lymph node (Fig. 4, 40) in the normal adult rabbit. *C* lymphocyte corona, *Cr* crypt, *E* epithelium, *G* germinal center, *I* interfollicular area, *P* paracortical area, *F* follicle, *M* medulla.

Figs. 3-4 Tonsil (Fig. 3, 40) and popliteal lymph

node (Fig. 4, 40) 6 days after the last whole-body irradiation of previously thymectomized animals. The difference in follicular repair between tonsil lymph node interfollicular areas and areas depleted of lymphocytes. *D* opening gland duct into the crypt. *GL* mucosa.





Figs 9-10 Tonsil (Fig. 9 $\times 130$) and popliteal lymph node (Fig. 10 $\times 60$) 3 days after the last whole-body irradiation with thymus shielding. Clear lymphocyte accumulations in the interfollicular areas of the tonsil (9) and the paracortical areas of the lymph node (10). Follicular structures (F) in the tonsil already reappearing (haematoxylin), as the lymph nodes absent.

Figs. 11-12 Tonsil (Fig. 11 $\times 130$) and lymph node (Fig. 12, $\times 80$) 6 days after the last whole body irradiation with thymus shielding. Large lymphocyte accumulations in the interfollicular areas of the tonsil (11) and the paracortical areas of the lymph node (12). F follicle.



Figs. 5-6. Tonsil (Fig. 5 $\times 40$) and popliteal lymph node (Fig. 6 $\times 50$) 4 weeks after the last whole-body irradiation of previously thymectomized animal. Normal follicular structures. Interfollicular areas and paracortical areas however without lymphocyte accumulations.

Figs. 7-8. Tonsil (Fig. 7 $\times 80$) and popliteal lymph node (Fig. 8 $\times 40$) 10 weeks after last whole-body irradiation of previously thymectomized animal. Absence of lymphocyte accumulations but normal follicular structures.

populations of lymphoid cells in the tonsil. The validity of the used experimental procedure can be concluded from our comparative findings in the popliteal lymph node. The observed separation of the two populations of lymphoid cells in this organ are in close agreement with those described by Keuning & van den Broek (1968), Nieuwenhuis et al. (1970) and Veldman (1970).

The conclusion is drawn that a thymus-dependent population of lymphoid cells is located in the well defined interfollicular areas of the tonsil. The follicular structures contain a second non-thymus-dependent population, temporarily eliminated by the irradiation procedure. About the origin of this latter population of lymphoid cells it is tentatively assumed that these cells originate from the bone marrow as has been demonstrated for the follicular structures of the popliteal lymph node and the spleen (Balner & Dersjant, 1964; Keuning & van den Broek, 1968; Veldman 1970).

According to these findings the tonsil seems to be fully comparable to the regional lymph nodes and the spleen. Although no definitive proof is given by these experiments, these two anatomically separate lymphoid cell populations in the tonsil can be supposed to correspond to the two different elements in the immunological system, the plasmacellular reaction and the cell-mediated immunity as has been demonstrated in the regional lymph nodes (Keuning & van den Broek, 1968; Nieuwenhuis et al., 1970; Veldman, 1970).

Confirmation of the immunological role of these two different cell populations by the application of suitable antigens, provoking a specific reaction in one of the systems, is rather difficult, since the tonsil is continuously exposed to antigenic stimulation. The presence of mast-type cells in the follicular structures and the interfollicular areas shortly after the total irradiation might be attributed to this stimulus antigen supply. In this way the temporal difference in time between the restoration of the follicular structures in the tonsil

and the lymph node is noticeable. It is likely that only a single secondary follicle is observed, which might be explained.

Apart from the total proof of the existence of a plasmacellular reaction in the tonsil, the data suggest the presence of a humoral component in the epithelium and the outer layer of the lymphoid tissue. Besides the support of this assumption can be derived from the experiments of Surján (1970) and Dierschke & Patt (1971). These authors have found a substantial production of bacterial antigens in the tonsillar tissue.

The above considerations lead to the conclusion that from birth onwards and a functional part of the adult animal derives from the regional lymph node the tonsil might play together with the lymphoid tissue, as is the first to be exposed to infection after birth.

ACKNOWLEDGEMENTS

The authors are greatly indebted to Professor Keuning and Dr Nieuwenhuis for their critical and valuable discussions, to Professor Kasten for his facilities provided and to Mrs T. Wilbert P. Spaan for their technical assistance.

ZUSAMMENFASSUNG

An normalen und thymektomierten Kaninchen wird mit Hilfe sublethaler Ganzkörperbestrahlung die Herkunft der lymphatischen Zellen in der Tonsille untersucht. Mit dieser Technik konnten zwei lymphoide Zellpopulationen verschiedener Herkunft nachgewiesen werden. Eine vom Thymus abhängige Population konnte in den interfollikulären Gebieten lokalisiert werden. Eine zweite wahrscheinlich vom Knochenmark abhängige Population zeigte sich in den follikulären Strukturen. Diese Beobachtungen scheinen die Behauptung zu rechtfertigen, dass es bei erwachsenen Tieren keine fundamentalen Unterschiede zwischen dem lymphatischen Gewebe der Tonsille und den regionalen Lymphknoten gibt.

REFERENCES

- Balber H. & Derjant, H. 1964 Early lymphatic regeneration in thymectomized radiation chimeras. *Nature* 204 941
- Fagrest, A. 1948 Antibody production in relation to the development of plasma cells. *Acta Med Scand*, Suppl. 204
- Fichtelius, K. E. 1968 Th. gut epithelium—a first level lymphoid organ? *Exp Cell Res* 49 87
- Godrick, E. A. & Patt, G. R. 1971 A comparison of the immune response of tonsils with the appendix and spleen in neonatal rabbits. *Acta Otolaryng* (Stockh.) 71 357
- Gowans, J. L., McGregor D. P. & Cowen, D. M. 1963 The role of small lymphocytes in the rejection of homografts of skin. In *The immunologically competent cell: its nature and origin*. Ciba Foundation Study Group no. 16 p. 20.
- Harrison, B. H. 1970. Tonsillar tissue implants in neonatally thymectomized mice. *J Immunol* 105 38.
- Keuning, F. J. 1965 Die Beantwortung eines homologen Gewebestimulus (Homotransplantation) durch das lymphoreticuläre System. *Oncologica* 19 180.
- Keuning, F. J. & van den Broek, A. A. 1968. Role of marginal zone cells of lymphoid follicles in the immune response of rabbits. *Exp Haematol* 17 43.
- Keuning, F. J., Dijkhuis, A. & Dijkstra-van der Vliet, Th. A. 1964 The effect of sublethal x-ray irradiation on the induction period of the antibody response in the rabbit. *Int J Radiat Biol* 8 279
- Meyer zum Gottesberge, A. & Loburg, E. 1967 Die Tonsille als Lymphozytöres Organ. *Acta Otolaryng* (Stockh.) 63 229
- Miller J. F. A. P. 1962. Role of the thymus in transplantation immunity *Ann NY Acad Sci* 99 340.
- Miller J. F. A. P. & Mitchell, G. F. 1968. Cell-to-cell interaction in the immune response. I. Hemolysin forming cells in neonatally thymectomized and reconstituted with thymus or thoracic duct lymphocytes. *J Exp Med* 128 801
- 1969 Thymus and antigen reactive cells. I. Antigen sensitive cells, their source and differentiation. *Transplantation Review* 1 Munksgaard Copenhagen.
- Nieuwenhuis, P., Veldman, J. E., van den Broek, A. A. & Keuning, F. J. 1970. Radiation induced dissociation of immune responsiveness in the rabbit. Delineation of a thymus derived and non-thymus derived lymphoid cell system. *Proc 1 Congr Int Radiobiol Physiochimie des Rayonnement* Eriks. Gordon & Breach Publ. London.
- Nossal, G. J. V., Cunningham, A., Mitchell, G. J. & Miller J. F. A. P. 1968. Cell to cell interaction in the immune response. III. Chromosomal mark analysis of single antibody-forming cells in reconstituted, irradiated or thymectomized mice. *J Exp Med* 128 839
- Oort, J. & Turk, J. L. A. 1965 A histological study of lymph nodes during the development of contact sensitivity in guinea pigs. *Brit J Exp Pathol* 46 147
- Sarjón, L. & Sarjón, M. 1970. The antibody production of tonsils. *Arch Otor Nas Kehlopphekk* 19 331
- Turk, J. L. & Oort, J. 1969 Further studies on the relation between germinal centres and cell-mediated injury. In: *Lymphatic tissue and peripheral in immune response*. *Adv Exp Med Biol* 5 317
- Veldman, J. E. 1970 *Histophysiology and microscopy of the immune response* Thesis. Groningen, The Netherlands.

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